When citing an abstract from the 2023 annual meeting, please use the format below.

[Authors]. [Abstract Title]. Program No. XXX.XX. 2023 Neuroscience Meeting Planner. Washington, D.C.: Society for Neuroscience, 2023. Online.

Poster

PSTR119. Cell Proliferation and Migration I

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR119.01/A1

Topic: A.01. Neurogenesis and Gliogenesis

Support: NIH Grant GR105748
NIH Grant GR107618
NIH Grant GR115824

Title: Interhemispheric Connectivity Fields in Olfactory Bulb and Piriform Cortex via Anterior Commissure

Authors: *E. MARTIN-LOPEZ[1], B. BRENNAN[2], N. SPENCE[2], C. A. GREER[3];
[1]Neurosurg., Yale Med. Sch., New Haven, CT; [2]Neurosurg., Yale Univ., New Haven, CT; [3]Dept Neurosci. & Neurosurg., Yale Univ. Dept. Neurosci. and Interdepartmental Neurosci. Program, New Haven, CT

Abstract: The anterior commissure (AC) is a bundle of axons that communicate interhemipherically between olfactory regions such as the olfactory bulb (OB), anterior olfactory nucleus (AON), and piriform cortex (PC) to integrate olfactory information bilaterally. Previously, we reported that the development of the AC is a highly regulated process that involves progressive and regressive strategies of growth, reaching the contralateral side at the end of the embryonic development at E17. Meanwhile, the arborization in the contralateral structures is delayed until postnatal days 3-5. Here, using adeno-associated viral (AAVs) vectors that transduce for EGFP or mCherry, we injected olfactory regions in the OB, AON, and PC to study the contralateral innervation fields traveling through the AC. We found that contralateral axons from the OB travel exclusively through the anterior limb of the AC to project into the granule cell layer (GCL). In contrast, axons originating in the anterior PC project into the contralateral OB, AON, and PC. These axons not only arborize into the GCL, but also into the mitral and external plexiform layers, and the anterior PC layer 1b. We observed the posterior PC projects exclusively to the contralateral posterior PC via the posterior limb of the AC, arborizing fundamentally in layer 1b. The endopiriform nucleus projects exclusively through the posterior limb of the AC towards posterior PC. Collectively, we show a detailed map of contralateral arborization within olfactory structures that may be critical to understanding the processing of olfactory information between the brain hemispheres.

Disclosures: E. Martin-Lopez: None. **B. Brennan:** None. **N. Spence:** None. **C.A. Greer:** None.

Poster

PSTR119. Cell Proliferation and Migration I

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR119.02/A2

Topic: A.01. Neurogenesis and Gliogenesis

Support: NIH Grant GR105748
NIH Grant GR107618
NIH Grant GR115824

Title: Lamination of embryonically-generated interneurons in the adult olfactory bulb

Authors: *N. SPENCE[1], K. HAN[2], E. MARTÍN LÓPEZ[3], B. BRENNAN[1], M. CHEVALLIER[2], C. A. GREER[4];
[1]Yale Univ., New Haven, CT; [3]Neurosurg., [2]Yale Med. Sch., New Haven, CT; [4]Dept Neurosci. & Neurosurg., Yale Univ. Dept. Neurosci. and Interdepartmental Neurosci. Program, New Haven, CT

Abstract: The olfactory bulb (OB) contains local circuits formed of excitatory mitral and tufted projection neurons (M/Tc). In addition, inhibitory interneurons, periglomerular neurons (PGNs) and granule cells (GCs) in the OB are critical for odor processing. These cells arrange in layers where the olfactory input is detected topographycally forming the OB odorant receptor map. Previously in the Greer Lab, we found that M/Tc are generated at different embryonic ages according to this map. Whether PGNs and GCs generated embryonically also follow a developmental pattern similar to that from M/Tc remains unknown. Here, using injections of thymidine analogs between the embryonic day 10 (E10) and E18, we tested the hypothesis that PGNs and GCs generated at different embryonic stages exhibit an age-dependent differential distribution in the OB. Our results show that interneurons located in the lateral regions of the OB are generated earlier compared to the medial regions, suggesting a lateral-to-medial developmental pattern similar to that seen in mitral cells. The analysis of thymidine analogs staining within the OB layers show that in the glomerular layer (GL), PGNs in the medial region were generated earlier than those in the lateral OB. This gradient was not seen in in the external plexiform, mitral and internal plexiform layers whose cells generated simultaneously. In contrast, in the granule cell layer, where the vast majority of OB interneurons are localized, we found that GCs in the lateral domains were generated earlier than those found in the medial domains, displaying an inverted gradient compared to the GL. While it is unclear if these developmental gradients in interneuron locations are connected to the developmental patterns previously reported for mitral cells, the results do underscore the importance of timing in the establishment of OB circuits. Building a better understanding of these neural circuits ultimately improves our ability to interpret disease states in which olfactory behaviors are altered.

Disclosures: **N. Spence:** None. **K. Han:** None. **E. Martín López:** None. **B. Brennan:** None. **M. Chevallier:** None. **C.A. Greer:** None.

Poster

PSTR119. Cell Proliferation and Migration I

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR119.03/A3

Topic: A.01. Neurogenesis and Gliogenesis

Support: NIH Grant GR105748
NIH Grant GR107618
NIH Grant GR115824

Title: Neurogenesis in the Anterior Olfactory Nucleus

Authors: *B. T. BRENNAN[1], E. MARTÍN LÓPEZ[2], M. CHEVALLIER[1], N. SPENCE[1], K. HAN[3], C. A. GREER[4];
[1]Yale Univ., New Haven, CT; [2]Neurosurg., [3]Yale Med. Sch., New Haven, CT; [4]Dept Neurosci. & Neurosurg., Yale Univ. Dept. Neurosci. and Interdepartmental Neurosci. Program, New Haven, CT

Abstract: The anterior olfactory nucleus (AON) is a laminar structure embedded within the olfactory peduncle that is heavily interconnected with the olfactory bulb (OB) and piriform cortex (PC). The main function attributed to the AON is a relay station between the OB and the superior processing centers of the brain in the PC and tubular striatum (formerly olfactory tubercle). The cytoarchitecture of the AON is made of a compact ring of projection neurons (*pars principalis*) that is subdivided into two parts. The outer plexiform layer (*opl*) thats contains axons coming from OB and PC, and the inner cell zone (*icz*) that is formed by the densely packed pyramidal cells. In comparison with other regions of the olfactory system, the development of the AON remains understudied. In this work, we injected thymidine analogs in pregnant mice between the embryonic day 10 (E10) to E18 to study the neurogenesis of the AON. We found that cells generated at E10 formed a ring between the OB axons and superficial *opl* (or layer 1a), as we previously reported in PC. Other *opl* cells were generated predominantly between E11-13. On the contrary, the neurons forming the *icz* were formed between E12-E14, coinciding with the generation of neurons in layer 1b of PC and in medial regions of the tubular striatum (Martin-Lopez et al, 2019a, b). For the first time, our data show a comprehensive timeline for the AON neurogenesis in mouse that is crucial to understanding the embryonic formation of the different relay stations along the olfactory pathway.

Disclosures: B.T. Brennan: None. **E. Martín López:** None. **M. Chevallier:** None. **N. Spence:** None. **K. Han:** None. **C.A. Greer:** None.

Poster

PSTR119. Cell Proliferation and Migration I

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR119.04/A4

Topic: A.01. Neurogenesis and Gliogenesis

Support: R01HD107489

Title: Gestational folic acid and/or vitamin B12 supply alter cortical neurodevelopment and interconnectivity in mouse offspring

Authors: *K. ZARBALIS[1], A. HARLAN DE CRESCENZO[2], L. TAT[2], N. CANNIZZARO[2], A. A. PANOUTSOPOULOS[3], Z. SCHAAF[3], S. RACHERLA[2], L. HENDERSON[2], K.-Y. LEUNG[4], N. D. E. GREENE[4], T. G. BOTTIGLIERI[5], R. GREEN[1];
[1]Pathology and Lab. Medicine/Shriners Hosp., [2]Dept. of Pathology and Lab. Med., [3]UC Davis, Sacramento, CA; [4]Inst. of Child Hlth., Univ. Col. London, London, United Kingdom; [5]Baylor Res. Institute, Metabolic Dis., Baylor Res. Institute, Metabolic Dis., Dallas, TX

Abstract: Folate is an essential micronutrient required for both cellular proliferation through *de novo* nucleotide synthesis and epigenetic regulation of gene expression through methylation. This dual requirement places a particular demand on folate availability during pregnancy when both rapid cell generation and programmed differentiation of maternal, extraembryonic, and embryonic/fetal tissues are required. Accordingly, folate in its synthetic form (folic acid, FA) is widely used in prenatal supplements and fortified foods to prevent neural tube defects and other congenital defects, a practice that has led to a substantial increase in the amount of FA intake during pregnancy in some populations. In addition, vitamin B12, deficiency can also adversely interfere with neurodevelopment through critically intersecting with the folate pathway and potentially exacerbating the effects of FA excess.In this study, we sought to define the neurodevelopmental consequences of imbalanced maternal folic acid and/or vitamin B12 supply during pregnancy in mouse offspring. We exposed groups of C57BL/6NJ mouse dams to amino acid defined rodent chow containing varying quantities of FA with or without concomitant lack of B12. Diets were initiated two weeks prior to breeding and maintained throughout pregnancy. Embryos were collected at embryonic day (E) 14.5 and pups on postnatal days (P) 0 and P21 to obtain brains for histological and biochemical analyses. In addition, brains of juveniles (P21) and young adults (P60) were processed for Golgi staining and subsequent Scholl analysis to reveal changes in neuronal complexity.Our results indicate that either maternal folate deficiency or FA excess in mice result in disruptions in folate metabolism of the offspring and paradoxically cause comparable changes in prenatal cerebral cortical neurogenesis by delaying early neurogenesis in favor of late-born neurons. These cytoarchitectural changes appear to have long-term effects by diminishing projection neuron dendritic arborization in either test group compared with controls. Similarly, B12 deficient offspring also exhibits changes in cortical development and neuronal complexity comparable to those of FA imbalance.In conclusions, our findings point to overlooked potential neurodevelopmental risks associated with excessively high levels of prenatal FA intake. In addition, they further illustrate adverse aspects of prenatal B12 deficiency and how the nutritional status of both vitamins is intertwined, creating the potential for deleterious outcome when perturbation of one or both micronutrients is disrupted.

Disclosures: K. Zarbalis: None. **A. Harlan De Crescenzo:** None. **L. Tat:** None. **N. Cannizzaro:** None. **A.A. Panoutsopoulos:** None. **Z. Schaaf:** None. **S. Racherla:** None. **L. Henderson:** None. **K. Leung:** None. **N.D.E. Greene:** None. **T.G. Bottiglieri:** None. **R. Green:** None.

Poster

PSTR119. Cell Proliferation and Migration I

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR119.05/A5

Topic: A.01. Neurogenesis and Gliogenesis

Support: National Key Research and Development Program of China (2020YFA0112700)
Shanghai Municipal Science and Technology Major Project (2018SHZDZX05)
the Strategic Priority Research Program of the Chinese Academy of Sciences (XDB32000000)
Ministry of Science and Technology of the People's Republic of China STI2030-Major Projects (2021ZD0204500)
the National Natural Science Foundation of China (31871035)
the State Key Laboratory of Neuroscience

Title: Homophilic interaction of cell adhesion molecule 3 coordinates retina neuroepithelial cell proliferation

Authors: *Y. LI, J. HE, B. XU, M. JIN, H. ZHANG, N. REN, J. HU;
Ctr. for Excellence in Brain Sci. and Intelligence Technology, Chinese Acad. of Sci., Shanghai, China

Abstract: Correct cell number generation is central to tissue development. However, in vivo roles of coordinated proliferation of individual neural progenitors in regulating cell numbers of developing neural tissues and the underlying molecular mechanism remain mostly elusive. Here, we showed that wild-type (WT) donor retinal progenitor cells (RPCs) generated significantly expanded clones in host retinae with G1-lengthening by *p15* (*cdkn2a/b*) overexpression ($p15^{+}$) in zebrafish. Further analysis showed that *cell adhesion molecule 3* (*cadm3*) was reduced in $p15^{+}$ host retinae, and overexpression of either full-length or ectodomains of Cadm3 in $p15^{+}$ host retinae markedly suppressed the clonal expansion of WT donor RPCs. Notably, WT donor RPCs in retinae with *cadm3* disruption recapitulated expanded clones that were found in $p15^{+}$ retinae. More strikingly, overexpression of Cadm3 without extracellular ig1 domain in RPCs resulted in expanded clones and increased retinal total cell number. Thus, homophilic interaction of Cadm3 provides an intercellular mechanism underlying coordinated cell proliferation to ensure cell number homeostasis of the developing neuroepithelia.

Disclosures: Y. Li: None. **J. He:** None. **B. Xu:** None. **M. Jin:** None. **H. Zhang:** None. **N. Ren:** None. **J. hu:** None.

Poster

PSTR119. Cell Proliferation and Migration I

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR119.06/A6

Topic: A.01. Neurogenesis and Gliogenesis

Support: NIH Grant EY007060
NIH Grant OD28612
NIH Grant EY007003
NIH Grant OD011021
Research to Prevent Blindness

Title: Il10 signaling governs the proliferation of Müller glia-derived progenitors via regulating inflammatory resolution of microglia during regeneration of photoreceptors in zebrafish

Authors: *M. NAGASHIMA, P. R. HAGAN, S. S. GARAPATI, T. HOANG, P. F. HITCHCOCK;
Univ. of Michigan, Ann Arbor, MI

Abstract: Inflammation is a universal tissue response to invasive pathogens and cellular injury. In the central nervous system, injury activates microglia to release inflammatory cytokines, a diverse group of soluble factors that function to activate, amplify, and resolve inflammation. Il10 is an anti-inflammatory cytokine that plays a critical role in resolving neuroinflammation. In the zebrafish retina, which has the capacity to regenerate neurons and photoreceptors, acute inflammation is required to reprogram Müller glia, the intrinsic stem cells, and regulate proliferation of the Müller glia-derived progenitors (MGDPs), the immediate antecedents of regenerated neurons. In this study we used wildtype and *il10*$^{-/-}$ mutants to determine the role of il10 signaling during regeneration of photoreceptors. Following photoreceptor death, in wildtype, expression of *il10* peaks at 2 days post lesion (dpl) and returns to baseline by 5 dpl. In the retinas, *il10 receptor alpha* is expressed exclusively by microglia. Q-PCR shows that pro-inflammatory cytokines, *tnfa*, *tnfb*, *il1b*, released from microglia, are significantly elevated in the loss of function mutants, *il10*$^{-/-}$, as compared with wildtype animals. In the mutant, activated microglia persist at the site of photoreceptor death. These results indicate that il10 signaling acts on microglia to facilitate inflammatory resolution during regeneration of photoreceptors. We next assayed proliferation of MGDPs and regeneration of photoreceptors in wildtype and *il10*$^{-/-}$ mutants. At 5 and 7 dpl in wildtype animals, MGDPs have exited the cell cycle and are differentiating into regenerated photoreceptors. In contrast, in *il10*$^{-/-}$ mutants, the MGDPs continue to proliferate. At 14 dpl in wildtype, regenerated photoreceptors have mature morphologies, whereas in the *il10*$^{-/-}$ mutants, photoreceptor morphology is markedly abnormal. These results indicate that in the zebrafish Il10 signaling governs the proliferation of MGDPs by determining the duration of microglial pro-inflammatory response. Further, we show that the persistent inflammation in *il10*$^{-/-}$ mutants compromises the maturation of regenerated photoreceptors.

Disclosures: M. Nagashima: None. **P.R. Hagan:** None. **S.S. Garapati:** None. **T. Hoang:** None. **P.F. Hitchcock:** None.

Poster

PSTR119. Cell Proliferation and Migration I

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR119.07/A7

Topic: A.01. Neurogenesis and Gliogenesis

Support: NIH Grant RF1MH12460501

Title: Brain-wide cellular resolution mapping of GABAergic cells and microglia unveils distinct spatiotemporal patterns in the developing mouse brain

Authors: *J. K. LIWANG[1], F. A. KRONMAN[1], Y.-T. WU[2], R. BETTY[1], J. A. MINTEER[1], D. SHIN[1], Y. KIM[1];
[1]Penn State Col. of Med., Hershey, PA; [2]Cedars Sinai Med. Ctr., Los Angeles, CA

Abstract: In the mammalian central nervous system, γ-aminobutyric acid-containing (GABAergic) neurons undergo embryonic neurogenesis and migration until programmed cell death occurs during early postnatal development, contributing to the establishment of the brain's inhibitory balance. Microglia, the brain's resident immune cells, also interact with GABAergic neurons during synaptic pruning and circuit maturation. Therefore, disruptions in this system are heavily implicated in neurodevelopmental conditions and disorders. However, our understanding of the typical developmental patterning of GABAergic cells and microglia in various brain regions is limited due to the lack of cellular resolution datasets during developmental periods. To bridge this knowledge gap, we established a multimodal Developmental Common Coordinate Framework (DevCCF) for the developing mouse brain at eleven time points (embryonic day (E) 11.5, E13.5, E15.5, E18.5, postnatal day (P) 4, P6, P8, P10, P12, and P14) using light sheet fluorescence microscopy and serial two-photon tomography. Subsequently, we employed quantitative cellular resolution mapping methods to chart and evaluate the growth of GABAergic cells during embryonic and early postnatal mouse brain development, including cell types expressing somatostatin (SST) and vasoactive intestinal peptide (VIP) during the early postnatal period. Additionally, we mapped early postnatal microglia throughout the entire brain. Our findings revealed spatiotemporal heterogeneity in the densities of both GABAergic and microglial cells across cortical and subcortical brain regions. The developmental trajectory of SST+ interneurons differed between sensory and association cortices, whereas VIP+ interneuron populations in cortical regions remained relatively stable. In contrast, microglial populations expanded proportionally with brain growth and demonstrated differential spatial distributions during early postnatal brain maturation. This study provides valuable insights into the region-specific development of inhibitory GABAergic circuits, including the involvement of microglia, in the developing mouse brain. Our aim is to utilize the cell type mapping methods employed in this project to characterize other diverse neuronal and non-neuronal cell types, as well as the cerebrovasculature, during early mouse brain development. Furthermore, establishing an open-access resource with the data from this project for the scientific community will be fundamental

in advancing our understanding of brain cell types and their roles in neurodevelopmental and neuropsychiatric disease processes.

Disclosures: **J.K. Liwang:** None. **F.A. Kronman:** None. **Y. Wu:** None. **R. Betty:** None. **J.A. Minteer:** None. **D. Shin:** None. **Y. Kim:** None.

Poster

PSTR119. Cell Proliferation and Migration I

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR119.08/A8

Topic: A.01. Neurogenesis and Gliogenesis

Support: Florida DOH 20K01
Jim and Betty Ann Rodgers Chair Fund

Title: Prenatal nicotine exposure via e-cigarettes reduces GABA cell density in the embryonic dorsal forebrain

Authors: *A. A. PARNELL[1], M. X. TRUPIANO[2], D. M. MCCARTHY[2], P. G. BHIDE[2]; [1]Biomed. Sci., Florida State Univ. Program In Neurosci., Tallahassee, FL; [2]Biomed. Sci., Florida State Univ., Tallahassee, FL

Abstract: E-cigarettes are marketed as a safer alternative to combustible cigarettes despite accumulating evidence to the contrary. The increase in the popularity of e-cigarettes among pregnant women necessitates an investigation into the impact of e-cigarettes on the developing fetus. The e-cigarette aerosol contains nicotine as well as a cocktail of chemicals (the e-liquid), which can produce its own adverse health effects. We used a whole-body aerosol exposure paradigm to examine the effects of e-cigarette (24mg/mL) and e-liquid aerosol on the development of the GABA neurons in the embryonic mouse brain. The e-liquid was a flavorless mixture of 50-50 propylene glycol and vegetable glycerin. Swiss Webster GAD67-GFP transgenic reporter mice were used to facilitate identification of GABA neurons based on GFP fluorescence in histological sections of the embryonic brain. Female mice were exposed to e-cigarette aerosol, e-liquid aerosol, or fresh air (control) for approximately 1.5 hours daily beginning 2 weeks prior to breeding and continuing through the 14th day of pregnancy. Neither the e-cigarette nor e-liquid exposure impacted dam bodyweight or embryo metrics. Embryos were removed on the 15th day and the number of GFP+ cells (GABA neurons) were calculated in the dorsal cerebral wall (future cerebral cortex) at two rostro-caudal levels of the forebrain. We found that e-cigarette aerosol exposure reduced the density of GABA neurons in the dorsal forebrain at the rostral ($F_{(2, 9)} = 6.09$, $p < 0.05$) but not caudal ($F_{(2, 9)} = 1.48$, $p > 0.05$) level. E-liquid alone did not produce significant effects at either level. Further, at the rostral level, e-cigarette aerosol exposure reduced GFP+ cell density in the dorsal ($p < 0.05$) and medial locations ($p < 0.05$) but not at lateral locations. These data suggest that exposure of the developing brain to e-cigarette aerosol produces deficits in GABA neuron numbers in the

developing cerebral cortex, and the effects appear to be due to the actions of nicotine rather than the e-liquid.

Disclosures: A.A. Parnell: None. **M.X. Trupiano:** None. **D.M. McCarthy:** None. **P.G. Bhide:** None.

Poster

PSTR119. Cell Proliferation and Migration I

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR119.09/A9

Topic: A.01. Neurogenesis and Gliogenesis

Support: R01MH099660
T32GM113896
T32NS082145
R25NS089462

Title: Tbx1, a transcription factor gene deleted in 22q11.2, is essential for myelination and cognitive speed in mice

Authors: *A. M. WELLS[1,2], T. HIRAMOTO[1], A. SUMIYOSHI[5,6], S. BOKU[7], Q. SHI[3], T. YAMAUCHI[1], K. TANIGAKI[8], G. KANG[1], R. RYOKE[5], H. NONAKA[5], M. BHAT[3], R. KAWASHIMA[5], N. HIROI[1,3,4];
[1]Pharmacol., [2]South Texas Med. Scientist Training Program, [3]Dept. of Cell. and Integrative Physiol., [4]Dept. of Cell Systems and Anat., Univ. of Texas Hlth. Sci. Ctr. At San Antonio, San Antonio, TX; [5]Inst. of Development, Aging, and Cancer, Tokohu Univ., Sendai, Japan; [6]Natl. Inst. for Quantum and Radiological Sci. and Technol., Chiba, Japan; [7]Neuropsychiatry, Kumamoto Univ., Kumamoto, Japan; [8]Shiga Med. Ctr., Shiga, Japan

Abstract: Cognitive deficits are debilitating impairments seen across neuropsychiatric disorders, some of which are thought to arise from aberrant structural processes. Copy number variations (CNVs) are relatively large genetic deletions or duplications that result in a wide spectrum of neuropsychiatric symptoms and cognitive deficits in humans. Carriers of 22q11.2 hemizygous deletions exhibit various cognitive deficits. Our previous studies showed that the heterozygous deletion of *Tbx1*, a transcription factor gene encoded within a 1.5 Mb commonly deleted segment of the 22q11.2 locus, slowed the speed to complete the Morris water maze and attentional set shifting task without slowing motor movement *per se*, and selectively reduced fractional anisotropy values and the number of large, myelinated axons in the fimbria in mice. As TBX1 protein is enriched in stem cells in post-embryonic mice, we hypothesized that TBX1 deficiency in this cell population contributes to the structural and cognitive deficits seen in 22q11.2 deletion syndrome. We examined the effects of conditional *Tbx1* heterozygosity in post-embryonic stem cells on cognitive functions one month later. When *Tbx1* heterozygosity was initiated in post-embryonic stem cells by tamoxifen given at postnatal day 1 to day 5 in nestinCreER^{TM};*Tbx1*$^{flox/+}$

(c*Tbx1*$^{+/-P1-P5}$) mice, they exhibited a slow speed to complete spontaneous alternation in a T-maze without slow motor movement; when *Tbx1* heterozygosity was initiated by tamoxifen at postnatal day 21 to day 25, there was no effect. To determine if TBX1 expression during the neonatal period is a sensitive window for stem cell development, work is in progress to determine the number of SOX2+/GFAP+ radial glia-like stem cells in zones of post-embryonic neurogenesis (e.g., subventricular and subgranular zones) of Tbx1 mutant and wild-type mice. Additionally, our ChIP-seq analysis showed that TBX1 binds to a locus near *FoxG1* among genes implicated in adult neurogenesis, cerebral dysmyelination, and neurodevelopmental disorders. Our data show that FOXG1 colocalizes with TBX1 and is expressed in subpopulations of SOX2+, DCX+, and NG2+ cells, markers for neural and oligodendrocyte progenitor cells. Our data suggest that TBX1 in neonatal, but not postnatal, stem cell populations may be required for the normal development of myelin in the fimbria and cognitive speed.

Disclosures: A.M. Wells: None. **T. Hiramoto:** None. **A. Sumiyoshi:** None. **S. Boku:** None. **Q. Shi:** None. **T. Yamauchi:** None. **K. Tanigaki:** None. **G. Kang:** None. **R. Ryoke:** None. **H. Nonaka:** None. **M. Bhat:** None. **R. Kawashima:** None. **N. Hiroi:** None.

Poster

PSTR119. Cell Proliferation and Migration I

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR119.10/Web Only

Topic: A.01. Neurogenesis and Gliogenesis

Support: INPER 2019-1-11

Title: Effect of maternal hyperglycemia on neuronal migration in the rat cerebral cortex: hypoactivity of reelin signaling

Authors: *R. VALLE-BAUTISTA, I. SOTO-VILLANUEVA, E. IDIAQUEZ-HERNÁNEZ, M. ORDOÑEZ-VILLANUEVA, N. DÍAZ-MARTÍNEZ, A. MOLINA-HERNÁNDEZ; Fisiología y Desarrollo Celular, Inst. Nacional de Perinatología, Ciudad de México, Mexico

Abstract: Gestational diabetes mellitus (GDM) is a condition that is characterized by presenting intolerance to carbohydrates for the first time during pregnancy. In Mexico, the prevalence of GDM in obese pregnant women is 13.7% (95% CI: 9.6 to 17.9), which is why it has become a public health problem. Some reports indicate that the children of diabetic mothers present cognitive deficiencies when they perform tasks that depend on the functional integrity of the cerebral cortex (Cx), for which it has been suggested that maternal hyperglycemia affects corticogenesis at the early stages of embryonic development. In previous studies, it was reported that in the rat hyperglycemia model, 14-day-old embryos (E14) and neonates on postnatal day 0 (P0) presented increased expression of MAP2 in the cortical neuroepithelium, which suggests an increase in neuronal differentiation and maturation. In addition, a mismatch was observed in the cortical layer markers, SATB2, FOXP2, and TBR1 at P21, suggesting alterations in the neuronal

migration process. In the present study, an evaluation of the cytoarchitecture of the Cx at P0 and P21 was carried out with H&E and Golgi-Cox staining, the expression and localization of reelin, and its receptors VLDLR and ApoER2, as well as N-cadherin (Ncad), were evaluated, using immunofluorescence in E16. The results show that the products of diabetic rats present pyramidal neurons invading the marginal zone at P0 and P21, in addition to a decrease in the reelin gradient and the expression of VLDLR, ApoER2, and Ncad at E16. Results of differential expression analysis of the neuroepithelium of diabetic rat embryos at E12 show that the reelin-mediated signaling pathway is hypoactive in the early stages of corticogenesis.

Disclosures: R. Valle-Bautista: None. **I. Soto-Villanueva:** None. **E. Idiaquez-Hernánez:** None. **M. Ordoñez-Villanueva:** None. **N. Díaz-Martínez:** None. **A. Molina-Hernández:** None.

Poster

PSTR119. Cell Proliferation and Migration I

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR119.11/A10

Topic: A.01. Neurogenesis and Gliogenesis

Support: NIH R01 NS088566
New York Stem Cell Foundation
National Science Foundation GRFP
Howard Hughes Medical Institute Gilliam Fellowship

Title: Signals Making a Splash: How the Choroid Plexus Harnesses Cerebrospinal Fluid for Developmental Communication

Authors: *Y. COURTNEY[1,2], E. YIMER[2], N. DANI[3], J. HEAD[4], M. LEHTINEN[2,1];
[1]Neurobio., Harvard Univ., Cambridge, MA; [2]Pathology, Boston Children's Hosp., Boston, MA; [3]Cell and Developmental Biol., Vanderbilt Univ., Nashville, TN; [4]Neurosci., Stanford Univ., Palo Alto, CA

Abstract: During embryonic brain development, cerebral cortical neurons form from neural progenitor cells that divide along the brain’s ventricles and in contact with cerebrospinal fluid (CSF). CSF is produced largely by the choroid plexus (ChP), an epithelial tissue in each ventricle whose cells secrete CSF and its contents, including instructive factors like IGF-2. The ChP also serves as a blood-CSF barrier, protecting the brain from infection and inflammation. Its cultivation of proper CSF composition is crucial to healthy development. Indeed, CSF aberrations are increasingly implicated in neurodevelopmental disorders including autism spectrum disorder (ASD), hydrocephalus, and schizophrenia. Despite its lifelong role in titrating CSF contents, ChP secretory machinery is poorly understood. Advances in imaging approaches enabled our discovery that, in addition to vesicular exocytosis, the ChP displays a high-capacity regulated exocytosis mechanism called apocrine secretion. We developed a toolbox using highly

expressed G-protein coupled serotonin receptor 5HT2C to evoke and interrogate apocrine secretion using standard biochemical approaches, multi-photon imaging, expansion microscopy, and electron microscopy. We demonstrate this mechanism's functionality during embryonic development, confirm that these events alter CSF composition in vivo, and report findings on their contents. We further provide evidence that this altering this secretion process negatively affects cortical brain development. This mechanism may be sensitive to stressors like maternal inflammation and environmental teratogens, with resulting perturbations leading to impaired brain development.

Disclosures: Y. Courtney: None. **E. Yimer:** None. **N. Dani:** None. **J. Head:** None. **M. Lehtinen:** None.

Poster

PSTR119. Cell Proliferation and Migration I

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR119.12/A11

Topic: A.01. Neurogenesis and Gliogenesis

Title: Protracted migration of immature neurons in the temporal lobe of young children

Authors: *S. L. SANTIAGO[1], M. A. NASCIMENTO[3], S. BIAGIOTTI[1], V. HERRANZ-PEREZ[4], R. BUENO[3], C. YE[3], T. ABEL[2], Z. ZHANG[5], J. RUBIO-MOLL[6], Z. YANG[5], J. GARCIA-VERDUGO[7], E. J. HUANG[8], A. ALVAREZ-BUYLLA[9], S. SORRELLS[1]; [1]Neurosci., [2]Neurolog. Surgery, Univ. of Pittsburgh, Pittsburgh, PA; [3]Univ. of California, San Francisco, San Francisco, CA; [4]Univ. de València, Valencia, Spain; [5]Fudan Univ., Shanghai, China; [6]Servicio de Obstetricia, Hosp. Universitari i Politècnic La Fe, Valencia, Spain; [7]Univ. Valencia, Valencia, Spain; [8]Dept Pathol, Univ. California- San Francisco, San Francisco, CA; [9]Dept Neurosurg., UCSF, San Francisco, CA

Abstract: The human medial temporal lobe contains the amygdala, entorhinal cortex (EC), and hippocampus, brain regions that are highly interconnected and are key for integrating emotional, spatial, and memory processing. These functions develop substantially in infancy, yet neurogenesis and neuronal migration to these regions are generally considered complete by birth. Here we find that the infant human temporal lobe contains many young neurons migrating into the amygdala, EC, and adjacent cortical regions until 2-3 years of age. These cells express markers of immature neurons (doublecortin, DCX) and inhibitory interneurons (distal-less homeobox 2, DLX2). We identified a focused stream of these neurons directed toward the EC that declined between birth and 11 months, resulting in individually migrating neurons in the EC until 2-3 years of age. Immunostaining and scRNA-seq of the migratory neurons and their postnatal targets revealed a progressive change in immature marker expression coinciding with their maturation. We find that the immature neurons migrating into the EC are almost entirely inhibitory LAMP5+ neurons, derived from the caudal ganglionic eminence (CGE). This investigation reveals a widespread migration of young inhibitory interneurons into multiple brain

regions in the human temporal lobe. This process contributes to excitatory-inhibitory balance and likely extends periods of plasticity within highly-interconnected brain circuits necessary for emotional, spatial, and memory processing.

Disclosures: **S.L. Santiago:** None. **M.A. Nascimento:** None. **S. Biagiotti:** None. **V. Herranz-Perez:** None. **R. Bueno:** None. **C. Ye:** C. Other Research Support (receipt of drugs, supplies, equipment or other in-kind support); Chan Zuckerberg Initiative, Chan Zuckerberg Biohub, Genentech. E. Ownership Interest (stock, stock options, royalty, receipt of intellectual property rights/patent holder, excluding diversified mutual funds); Related Sciences, ImmunAI, Maze Therapeutics. F. Consulting Fees (e.g., advisory boards); Related Sciences, ImmunAI, Maze Therapeutics, TReX Bio.. **T. Abel:** None. **Z. Zhang:** None. **J. Rubio-Moll:** None. **Z. Yang:** None. **J. Garcia-Verdugo:** None. **E.J. Huang:** None. **A. Alvarez-Buylla:** E. Ownership Interest (stock, stock options, royalty, receipt of intellectual property rights/patent holder, excluding diversified mutual funds); Neurona Therapeutics. F. Consulting Fees (e.g., advisory boards); Neurona Therapeutics. **S. Sorrells:** None.

Poster

PSTR119. Cell Proliferation and Migration I

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR119.13/A12

Topic: A.01. Neurogenesis and Gliogenesis

Support: NINDS Grant R01NS116054

Title: Genetic mosaic analysis of abscission genes Cep55 and Kif20b in the developing cortex

Authors: *H. DINGSDALE[1], K. FILIPEK[1], N. DWYER[2];
[1]Cell Biol., Univ. of Virginia, Charlottesville, VA; [2]Univ. of Virginia Sch. of Med., Sch. of Med., Charlottesville, VA

Abstract: Tight regulation of neural stem cell (NSC) division in the developing cortex is crucial for generating the correct number and placement of neurons and glia. Mitosis and cytokinesis of NSCs occur at the ventricular surface, and mediate the segregation of cell fate determinants. The location and duration of abscission, the severing of the last connection between the mother and daughter cells, is tightly controlled. Our previous data show that mutations that either delay or accelerate NSC abscission have drastic effects on cortical development. Deletion of the coiled-coil protein Cep55 in mice leads to **longer** abscission times, with some failed cell divisions, apoptosis, and microcephaly. Mutation of Kif20b, a kinesin motor protein, causes **shorter** abscission times, but this also leads to increased apoptosis and microcephaly. Furthermore, Kif20b has a post-mitotic role in regulating the neuronal microtubule cytoskeleton, axon outgrowth, and branching. In the germline mutant brains, the abnormal brain structure and numerous apoptotic cells make it difficult to isolate cell autonomous from non-autonomous effects. To overcome this, here we use Mosaic Analysis of Double Markers (MADM) to generate

fluorescently tagged sparse knockout cells within a normal-sized brain. Upon Cre-mediated recombination, division of a mother NSC generates one EGFP-expressing knockout (green), and one tdTomato-expressing wildtype (WT; red) daughter cell, the latter of which serves as a convenient internal control. MADM-labelled WT littermates, in which both red and green cells are WT, serve as additional controls. By crossing MADM-19 mice to *Cep55* and *Kif20b* mutant lines and isolating recombinants, we successfully generated both MADM-*Cep55* and MADM-*Kif20b* mouse lines. Using the Emx1-Cre line to drive recombination specifically in the cortex starting at embryonic day (E)10.5, MADM-*Cep55* brains show easily traceable labelling of both red and green cells. In control brains, there were approximately equal numbers of red (*Cep55* WT) and green (*Cep55* WT) cells at postnatal day 2. In experimental cortices however, there were half as many green (*Cep55* knockout) cells compared to red (*Cep55* WT) cells, demonstrating that *Cep55* loss disrupts neurogenesis. We examined changes earlier in development at E16 and E12, assessing the autonomous and non-cell autonomous functions of Cep55 and Kif20b on cell morphologies, cell migration, and cell fates. These analyses will significantly improve our understanding of the complex cell interactions occurring during cortical development, and both the short- and long-term consequences of disrupted cell division.

Disclosures: **H. Dingsdale:** None. **K. Filipek:** None. **N. Dwyer:** None.

Poster

PSTR119. Cell Proliferation and Migration I

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR119.14/A13

Topic: A.01. Neurogenesis and Gliogenesis

Support: FONDECYT #1221376 (to DB)
FONDECYT #1211384 (to FB)
Basal funding for Scientific and Technological Center of Excellence, IMPACT, #FB210024 (to DB and FB)
Ph.D. fellowship ANID #21201204 (to FS)

Title: Exploring the potential link between vitamin E deficiency and adverse neurodevelopment in fetuses lacking the HDL receptor SR-B1

Authors: *F. SAAVEDRA[1], N. G. SANTANDER[2], M. MENDEZ[1], M. T. ORELLANA[1], L. BATIZ[1], D. BUSSO[1];
[1]Univ. de los Andes, Santiago, Chile; [2]Inst. de Ciencias de la Salud, Univ. de O´Higgins, Rancagua, Chile

Abstract: During neurogenesis, neural stem cells proliferate and differentiate as they migrate from the ventricular (VZ) and subventricular (SVZ) zones to the brain cortex. These processes, which begin during early gestation (at gestational day (E) 10.5 in mice) and continue postnatally, are hindered by gestational exposures, including nutritional deficiencies. Vitamin E (VitE) is a

lipophilic micronutrient with antioxidant and non-antioxidant functions. Gestational VitE deficiency increases fetal oxidative stress and affects offspring neurodevelopment and postnatal behavior. VitE is taken up by the embryo from maternal plasma through lipoprotein receptors in extraembryonic tissues. The HDL receptor Scavenger receptor class B type 1 (SR-B1) is expressed in placenta and blood-brain barrier cells and can take up VitE. We hypothesized that SR-B1$^{-/-}$ fetuses exhibited brain VitE deficiency and altered brain morphometry and neurogenesis. SR-B1$^{-/-}$ and WT fetuses at E16.5 were generated by heterozygous intercrossing. VitE content was evaluated in SR-B1-/- and WT fetal brains, placenta, and plasma by HPLC. Lipoperoxidation was determined using TBARS. Coronal sections of SR-B1$^{-/-}$ and WT fetal brains were stained with H&E and subjected to morphometric analyses or used to characterize neurogenesis through the presence and localization of markers for specific cell types using indirect immunofluorescence (IIF). Cell proliferation was evaluated using anti-Ki67 in IIF. Our results showed that VitE levels were similar in placenta and fetal serum from both genotypes. However, significantly lower VitE concentrations were found in SR-B1$^{-/-}$ fetal brains (305 vs. 1074 pg/µg protein in WT, p=0.021, n=8) and livers (27.2 vs. 121.1 pg/µg protein in WT, p=0.06, n=7). Interestingly, SR-B1$^{-/-}$ fetal brains showed increased lipoperoxidation (25.69 vs 18.33 nmol MDA/mg protein in WT, p=0.04, n=7) and lower proliferation rates in the SVZ (0.05 vs 0.13 KI67^{+}/total cells, p=0.004, n=3). We did not detect gross differences in ventricle/hemisphere areas ratio or cortical thickness. These results suggest that low VitE levels are associated with increased lipoperoxidation and deficient neural stem cell proliferation in SR-B1$^{-/-}$ fetal brains. This model may provide valuable insight into the mechanisms underlying neurodevelopmental derangements due to VitE deficiency.

Disclosures: **F. Saavedra:** None. **N.G. Santander:** None. **M. Mendez:** None. **M.T. Orellana:** None. **L. Batiz:** None. **D. Busso:** None.

Poster

PSTR119. Cell Proliferation and Migration I

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR119.15/A14

Topic: A.01. Neurogenesis and Gliogenesis

Support: NIH Grant U18DA052402
NIH Grant R01HL139492

Title: A novel neuroimmunomodulatory cocktail or TLR4KO repairs the developing brain from injury caused by prenatal methadone exposure

Authors: *V. OMONIYI[1,2], N. MADURAI[2], Y. KITASE[2], E. CHIN[5], S. ROBINSON[3,4], L. JANTZIE[2,3,5];
[1]Neonatal-Perinatal Med., Johns Hopkins Med. Institutions, Baltimore, MD; [2]Pediatrics, [3]Neurosurg., [4]Neurol., Johns Hopkins Univ. Sch. of Med., Baltimore, MD; [5]Kennedy Krieger Inst., Baltimore, MD

Abstract: Substance misuse during pregnancy and its impact on postnatal outcomes is a critical threat to pediatric and adult health. An urgent need exists to define the full spectrum of sequelae associated with prenatal opioid exposure (POE), including long-term deficits persisting beyond withdrawal in the neonatal period. Here, we tested the hypothesis that a cocktail designed for neurorepair, erythropoietin plus melatonin (EPO+MLT), would protect the developing brain against the long-term structural and functional deficits associated with POE. We also defined the inflammatory burden and role of Toll-Like receptors-4 (TLR4s) in impaired neurodevelopment induced by methadone. Wild-type (WT) or TLR4KO pregnant rats were randomized to osmotic mini pumps containing saline or methadone (12 mg/kg, 0.25 µL/hour flow rate for 28 days). On P1, male and female WT pups were randomized to receive EPO (2500U/kg from P1-P5) and MLT (20mg/kg from P1-P10) or saline treatment. Gait in adulthood, attention, cognition, white matter microstructure and functional activation were assayed in adulthood by blinded observers. Differences were examined using two-way ANOVA with Bonferroni correction ($p<0.05$; n=10-15/group). Methadone induced a TLR4-dependent immune response that resulted in upregulation of pro-inflammatory mediators, concomitant with behavioral disinhibition, hyperactivity, and hypermobility ($p<0.001$ for all). TLR4KO attenuated methadone-induced serum increases in IL-1, CXCL1, TNF and IL-6 ($p<0.01$ for all) associated with reversal of hyperactivity and disinhibition ($p<0.05$). Treatment with EPO+MLT reversed methadone induced gait deficits, disinhibition, and mitigated deficits of attention on a touchscreen task ($p<0.01$). Multimodal neuroimaging revealed methadone-induced reductions in fractional anisotropy in major white matter tracts ($p<0.001$), as well as diminished brainwide functional connectivity ($p<10^{-6}$). Decreases in cortical-thalamic and thalamo-basal ganglia connectivity were particularly prominent with large effect sizes (Glass's $\Delta>1$) and these were also attenuated by EPO+MLT. Taken together, adult animals exposed to methadone prenatally have profound deficits of attention, gait, activity, inhibition and inflammation. These functional deficits co-occur with deficits in structural and functional connectivity. Mechanistically targeted neuroimmunomodulation with EPO+MLT or TLR4KO both protect the developing brain from the detrimental effects of methadone. This therapeutic response indicates novel treatment options for the countless children with POE born each year.

Disclosures: V. Omoniyi: None. **N. Madurai:** None. **Y. Kitase:** None. **E. Chin:** None. **S. Robinson:** None. **L. Jantzie:** None.

Poster

PSTR119. Cell Proliferation and Migration I

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR119.16/A15

Topic: A.01. Neurogenesis and Gliogenesis

Title: Investigating the role of dysregulated metabolism in pediatric glioblastoma

Authors: *A. WIESA, S. MCGEE, R. SAMARASINGHE;
Sch. of Med., Deakin Univ., Melbourne, Australia

Abstract: Pediatric glioblastoma (pGBM) is an aggressive brain tumour with low survival rates. Traditional treatments for adult glioblastoma have limited effectiveness in pGBM. Unique biological features of pGBM necessitate targeted therapies. Aberrant glucose metabolism, specifically enhanced glycolysis, and lactate production (the Warburg effect) is prominent in adult GBM. This metabolic shift drives cancer cell proliferation, chemoresistance, rapid growth, and metastasis. While the role of glucose metabolism in adult GBM has been extensively researched, the investigation into metabolic dysregulation within pGBM remains an open field of exploration. We sought to understand glucose metabolism in pGBM (SF188) and adult GBM (T98G) and compare it to non-cancerous cells (CT003 and HEK293) in both monolayer and 3D spheroids; with the hypothesis that the cancers exhibit dysregulated metabolism. We assessed the metabolic phenotype of these cells using the Seahorse XF analyzer, measuring both oxidative and glycolytic metabolic flux. All data were normalized based on protein concentration (BCA). The pGBM and T98G cells exhibited significantly higher rates of oxygen consumption (OCR) measured at 2.7 ± 0.21pmol/min/µg protein and 4.5 ± 0.24pmol/min/µg protein, respectively. They also displayed higher extracellular acidification rates (ECAR) at 0.98 ± 0.04mpH/min/µg protein and 1.4 ± 0.04mpH/min/µg protein, respectively. These rates were observed to be considerably higher when compared with those of the CT003 and HEK cell lines, which exhibited OCR values of 0.91 ± 0.11pmol/min/µg protein and 0.91 ± 0.13pmol/min/µg protein, and ECAR values of 0.30 ± 0.02mpH/min/µg protein and 0.1 ± 0.02mpH/min/µg protein, respectively (n=4 ± SEM). Our research indicates that cancer cells exhibit heightened glycolytic and aerobic metabolism, thus signifying an increased energy capacity. This is particularly noticeable in the metabolic activity difference between adult and pediatric GBM cells in both 3D models and monolayer contexts. Notably, pediatric GBM (pGBM) metabolism aligns with the Warburg effect and differs from its adult equivalent, prompting a tailored strategy for future research into these distinct diseases. Further studies will focus on understanding the role of elevated glycolysis in pGBM and how this metabolic shift influences its proliferation and survival. This could provide insights for developing therapies specifically targeting pGBM metabolism.

Disclosures: **A. Wiesa:** None. **S. McGee:** None. **R. Samarasinghe:** None.

Poster

PSTR119. Cell Proliferation and Migration I

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR119.17/A16

Topic: A.01. Neurogenesis and Gliogenesis

Support: Hamilton College

Title: Identification and Characterization of a Tyramine Beta-Hydroxylase-Related Gene Product in Drosophila melanogaster.

Authors: *H. K. LEHMAN, A. CHOI;
Biol. and Neurosci., Hamilton Col., Clinton, NY

Abstract: Dopamine β-hydroxylase (Dbh), tyramine β-hydroxylase (Tbh), and peptidyl-glycine alpha-amidating monooxygenase are type II ascorbate-dependent monooxygenases, a class of hydroxylating enzymes that require copper as a cofactor, ascorbate as an electron donor, and oxidize substrates. These monooxygenases have different substrate specificities; nevertheless, there is considerable sequence similarity amongst the enzymes. Other putative enzymes similar, but not identical to Dbh and Tbh have been recently described. Human monooxygenase X (MOXD1) encodes a protein 25% similar to human Dbh, and Drosophila MOXD1 (dMOXD1) is 32% identical to Drosophila Tbh. The tissue distribution of Dbh also differs substantially from MOXD1. For example, Dbh is restricted to adrenal glands, pons, liver, midbrain, medulla oblongata, whereas MOXD1 is much more broadly expressed (smooth muscles, endometrium, seminal vesicles, kidney, adrenal glands, prostate, gallbladder, salivary glands, cerebral cortex, hippocampus, and basal ganglion) (Human Protein Atlas, 2023). Thus, Dbh and MOXD1 have unique distributions suggesting that MOXD1 has a novel substrate and function. However, despite several studies and the broad distribution of MOXD1, the endogenous substrate(s) for this putative hydroxylase enzyme has not been identified and the function of this widely distributed protein is enigmatic.
The structural and enzymatic similarities between Dbh and Tbh suggest that an ortholog to MOXD1 should also be present in insects. Indeed, Xin et al. (2004), noted that monooxygenase family members are present in Drosophila melanogaster and Caenorhabditis elegans. We have investigated the distribution of dMOXD1 by examining the its expression and localization. PCR and in situ hybridization studies indicate that dMOXD1 is highly expressed in Drosophila early embryos, larval brain, and adult brain. Expression is early embryos is most abundant in early (1-3 hr) and mid stage (11-12 hr) neuroblasts. To examine the subcellular distribution of dMOXD1, NIH3T3 cells were transfected with pAcGFP-N1 vectors encoding a dMOXD1-GFP fusion protein and was colocalized with protein dismutase isomerase to demonstrate that dMOXD1is restricted to the endoplasmic reticulum. Further, endogenous dMOXD1 mRNA is upregulated in Drosophila S2 cells under hypoxic (2% O_2) and ER stress (2 mM dTT) conditions. Current studies are focused on the link between dMOXD1 and ER stress. In sum, our study provides evidence that dMOXD1 is structurally similar to Tbh, it is strongly expressed during early development and in the mature nervous system of Drosophila, and its expression is influenced by cellular stress.

Disclosures: H.K. Lehman: None. **A. Choi:** None.

Poster

PSTR119. Cell Proliferation and Migration I

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR119.18/A17

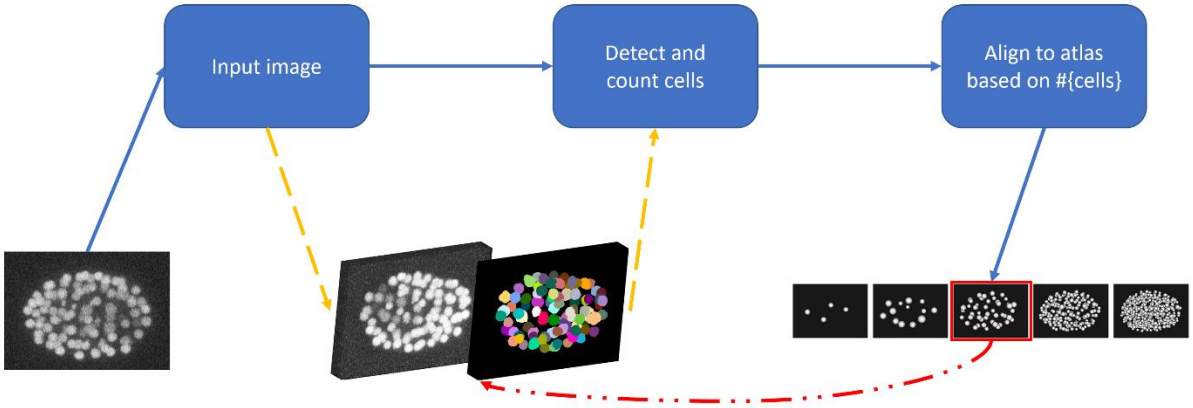
Topic: A.01. Neurogenesis and Gliogenesis

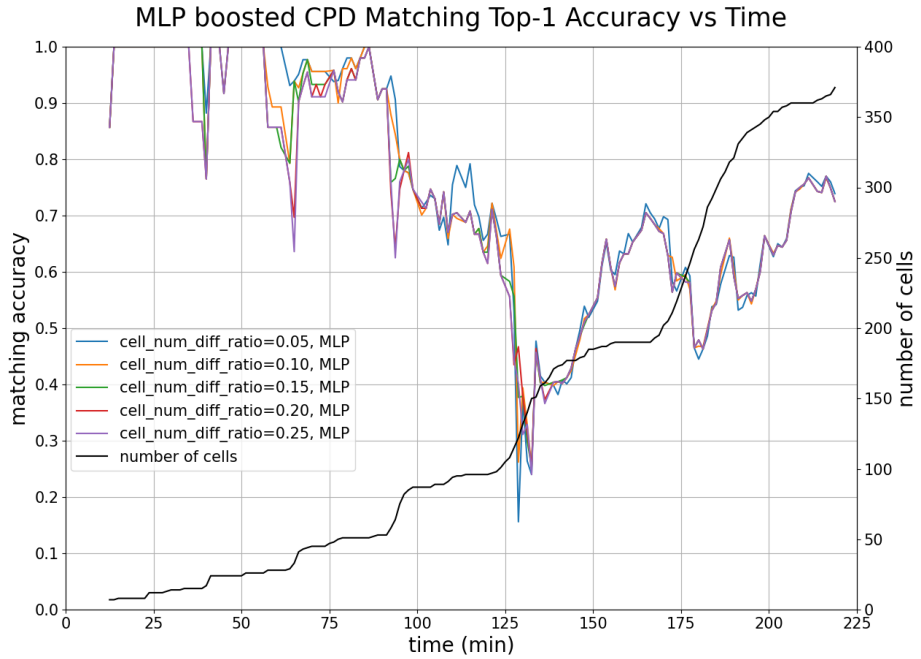
Support: GBMF8815

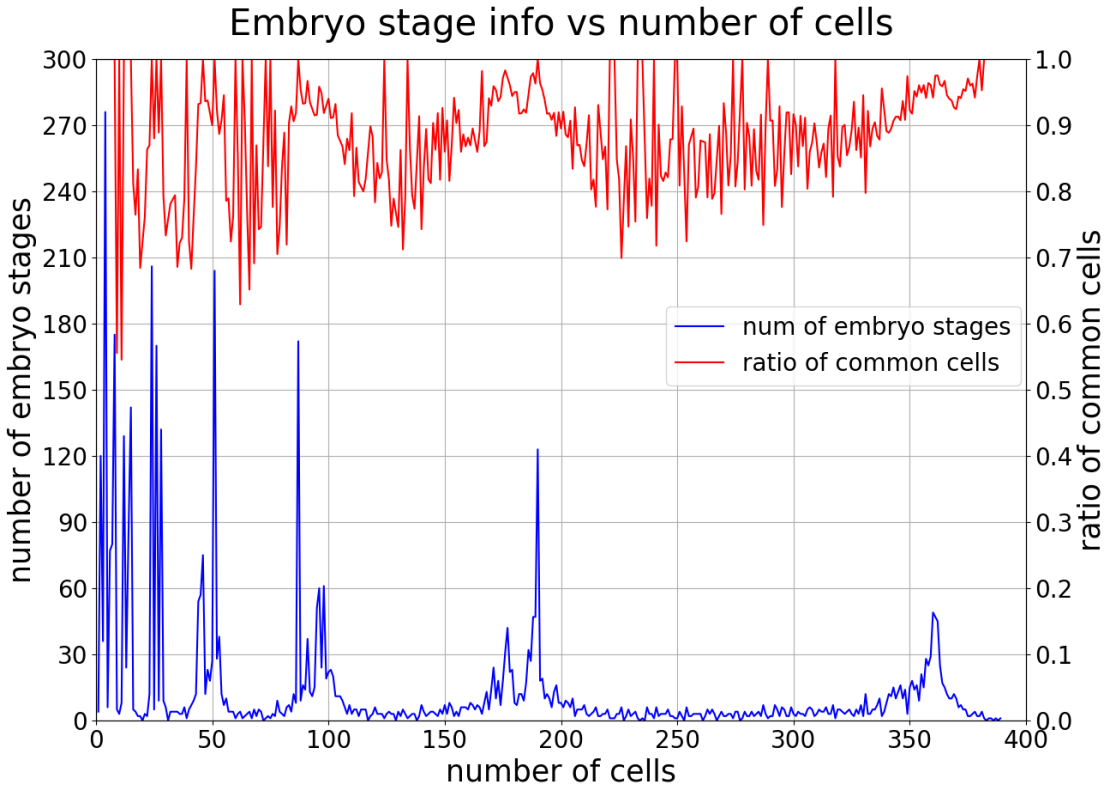
Title: Atlas is all you need

Authors: *B. DAI[1], G. KROESCHELL[2], R. CHRISTENSEN[2], A. SANTELLA[3], H. SHROFF[2], P. LA RIVIERE[1];
[1]Univ. of Chicago, Chicago, IL; [2]Janelia Res. Campus, Howard Hughes Med. Inst. (HHMI), Ashburn, VA; [3]Mem. Sloan Kettering Cancer Ctr., New York City, NY

Abstract: Identifying cells in embryo development with low temporal and spatial resolution time-lapse imaging is difficult. The general solution is cell tracking, where cells are identified by processing the whole sequence of images and comparing with the lineage. Here, by leveraging the invariant lineage of *C. elegans*, we developed the first atlas based approach for cell identification in worm embryos. Cells are identified by matching against the dynamic atlas with point set registration. Using only cell positional information, our method reached >77% accuracy at 371 cell stage with the help of machine classification. The method requires only the current frame of image and can output results within a minute, which greatly reduced the running time compared with cell tracking. We anticipate the method would be used in real-time single-cell experiments when multicolor strain is available for worm embryos in the near future.







Disclosures: **B. Dai:** A. Employment/Salary (full or part-time); University of Chicago.

Poster

PSTR120. Neural Stem Cells and Differentiation

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR120.01/A18

Topic: A.01. Neurogenesis and Gliogenesis

Support: NIH Grant R01NS123263

Title: Mapping differentiation patterns of human radial glia using single cell lineage tracing

Authors: *M. STEYERT[1], M. KEEFE[1], R. DELGADO[3], D. SHIN[4], C. KIM[2], T. NOWAKOWSKI[1];
[2]Dept. of Anat., [1]Univ. of California San Francisco, San Francisco, CA; [3]Neurosurg., Harvard Med. Sch., Boston, MA; [4]Anatomy; Psychiatry, Univ. of California, San Fransisco, San Francisco, CA

Abstract: The variety of human neocortical cell types originate from progenitors known as radial glia that span the cortical wall and serve as the stem cells of the cortex. Midway through neurogenesis, this uniform population splits into truncated radial glia that reside in the ventricular zone (VZ) and outer radial glia that are located in the outer subventricular zone (OSVZ). Expansion of progenitor pools in humans and other large-brained mammals compared to mice raises questions about the roles of these unique stem cell niches. To understand the lineage potential of these radial glia subtypes, we used a novel lineage tracing tool called STICR that marks individual cells with a highly diverse, heritable DNA barcode. We report that both germinal zones are capable of generating the full diversity of cell types. Interestingly, OSVZ progenitors are biased toward the generation of cortical-born interneurons, which have only recently been described in humans and have not been identified in mice. Surprisingly, VZ progenitors generate a large share of oligodendrocytes, and continue to generate deep-layer neurons late into neurogenesis. This suggests a potential new source for subplate neurons in humans, and underscores the importance of truncated radial glia in excitatory neurogenesis. Together, these findings shed light on the roles for the VZ and OSVZ niches in human cortical development.

Disclosures: M. Steyert: None. **M. Keefe:** None. **R. Delgado:** None. **D. Shin:** None. **C. Kim:** None. **T. Nowakowski:** None.

Poster

PSTR120. Neural Stem Cells and Differentiation

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR120.02/A19

Topic: A.01. Neurogenesis and Gliogenesis

Support: DBT/Wellcome Trust India Alliance: Intermediate Career Fellowship (IA/I/19/1/504288)
DST-SERB (SRG/2020/001612)
DBT- Har Gobind Khorana Innovative Young Biotechnologist Fellowship (BT/13/IYBA/2020/02)

Title: Lysine specific demethylase 1 (LSD1) regulates neurogenesis in human neural stem cells by repressing human neural stem cell-enriched genes

Authors: *A. CHANNAKKAR[1,2], L. D'SOUZA[1], A. KUMAR[1], K. KALIA[1], K. PHALNIKAR[1], P. CHANDRAMOULI REDDY[3], B. MURALIDHARAN[1];
[1]Inst. for Stem Cell Sci. and Regenerative Med., Bengaluru, India; [2]Regional centre for biotechnology, Faridabad, India; [3]Dept. of Life Sci., Shiv Nadar Inst. of Eminence, Delhi NCR, India

Abstract: The cerebral cortex is the seat of all higher order functions in the brain namely, sensory perception, decision-making, language, learning and memory. Dynamic modulation of chromatin state throughout the development regulates the precise and coordinated orchestration of neural progenitor proliferation, differentiation, migration and maturation of neurons to form functional circuitry. Lysine specific demethylase LSD1 removes methyl groups from mono and di-methylated lysine on Histone 3 (H3K4me1/2 and H3K9me1/2). It is a part of the chromatin remodeler complex NuRD (where it acts by its demethylase activity) and CoREST. LSD1 has been reported to function differentially between mice and humans, it positively regulates progenitor proliferation in mice, whereas, it promotes neuronal differentiation in humans. However, downstream targets and regulatory mechanism of LSD1 in human neuronal development are not clear. We have performed LSD1 ChIP-seq and RNA-seq and Histone ChIP seq upon LSD1 inhibition in human neural stem cells (NSCs) to ascertain its genome-wide occupancy. Our study revealed that LSD1 regulates human neuronal development via repression of human brain enriched/specific downstream effector genes crucial for cell-cell communication and signalling. Transition from Neuroepithelial cells to Radial Glia cells is dependent on Notch signalling. This signalling is also essential for progenitor pool maintenance. We report that on LSD1 represses the expression of Notch signalling genes such as TLE1 and HES6. Overexpression of these genes in human NSCs phenocopies LSD1 inhibition phenotype. We have further gone onto characterise the role of other key novel downstream effector in regulating human neuronal development.

Disclosures: A. Channakkar: None. **L. D'Souza:** None. **A. Kumar:** None. **K. Kalia:** None. **K. Phalnikar:** None. **P. Chandramouli Reddy:** None. **B. Muralidharan:** None.

Poster

PSTR120. Neural Stem Cells and Differentiation

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR120.03/A20

Topic: A.01. Neurogenesis and Gliogenesis

Title: An early role in human cortical development for the disease associated CNTNAP2

Authors: M.-Y. LI[1], X. CHEN[1], S.-J. YOON[1], S. KANTON[1], K. W. KELLEY[1], A. M. VALENCIA[1], L. BICKS[2], Q. GUO[2], L. LI[1], M. THETE[1], A. M. PASCA[1], J. R. HUGUENARD[1], D. H. GESCHWIND[2], *S. P. PASCA[1];
[1]Stanford Univ., Stanford, CA; [2]UCLA, Los Angeles, CA

Abstract: Human cerebral cortical development is a protracted process that involves cell specification, migration, synaptogenesis and maturation. We surprisingly found that the neurexin superfamily member CNTNAP2, whose bi-allelic loss is associated with severe intellectual disability, language regression and early onset epilepsy is surprisingly expressed in newborn immature cortical glutamatergic neurons. Using stem cell-derived human cortical organoids (hCOs), we discovered highly synchronous spontaneous calcium activity following CNTNAP2 knockout. Similarly, acute application of a monoclonal antibody targeting the extracellular domain of CNTNAP2 was sufficient to induce a hypersynchronous phenotype in control hCOs. Pharmacological studies revealed that this defect is related to spiking activity and NMDA receptors. Functional studies further indicated that loss of CNTNAP2 is associated with increased, non-synaptic, glutamate bulk transmission, and suggested that this adhesion molecule may function as a ‘brake’ on neural activity at early stages of cortical development. To further explore this, we used an ex vivo human primary tissue preparation. Live imaging in organotypic slices showed that spontaneous calcium activity increased from the frontal to the occipital cortex, which was inversely correlated with CNTNAP2 gene expression. Application of a CNTNAP2 monoclonal antibody increased activity in frontal slices to parietal cortex levels. Taken together, these results suggest an early developmental, non-synaptic role in human cortical neuronal maturation for a synaptic cell adhesion molecule associated with severe disease.

Disclosures: M. Li: None. **X. chen:** None. **S. Yoon:** None. **S. Kanton:** None. **K.W. Kelley:** None. **A.M. Valencia:** None. **L. Bicks:** None. **Q. Guo:** None. **L. Li:** None. **M. Thete:** None. **A.M. Pasca:** None. **J.R. Huguenard:** None. **D.H. Geschwind:** None. **S.P. Pasca:** None.

Poster

PSTR120. Neural Stem Cells and Differentiation

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR120.04/A21

Topic: A.01. Neurogenesis and Gliogenesis

Title: Determining the local circuit dynamics of lineage-based neurons in the mouse neocortex

Authors: A. HORVATH[1], W. A. TYLER[1], *H. QADIR[2], T. HAYDAR[1];
[2]Ctr. for Neurosci., [1]Children's Natl. Hosp., District of Columbia, DC

Abstract: The most recently evolved part of the mammalian brain is the 6-layered neocortex. Developed in early gestation, this diverse network of complex circuitry is responsible for higher order functioning such as consciousness, attention, and memory. The expansion of the neocortex occurs via two distinct neuronal lineages- those that differentiate directly from apical radial glial cells at the ventricular zone and those that undergo indirect neurogenesis via an intermediate progenitor cell. The distinct morphological, electrophysiological, and transcriptomic diversity between neurons arising from each lineage has been described, but how it contributes to the circuitry within the mature neocortex is unknown. Further understanding of why these neuronal lineages are produced may provide insight to disorders with neuron production deficits such as Down Syndrome and Schizophrenia, where alterations in neurogenesis may result in changes to cortical function.
To probe whether different neuronal lineages have preferential connectivity in the postnatal neocortex, we developed a paradigm to examine lineage-specific microcircuitry using 2-Photon microscopy. We first performed *in utero* electroporation in the fetal mouse neocortex at embryonic day 14.5 to fluorescently label precursor cells that produce layer 2/3 pyramidal neurons of the neocortex. This fate mapping DNA construct also encodes a calcium reporter (JyCamP) to be expressed by all cells and a photoactivateable ion channel (channelrhodopsin-2) in one of the two lineages to allow for optical stimulation during *in vivo* imaging. When the neocortex is fully mature after postnatal day 21, a cranial window was placed at the site of transfection to examine the neurons produced from the previously targeted progenitor cells. Neurons are visualized using 2-Photon laser-scanning microscopy to induce and observe calcium fluctuations in the intact brain. Here, we recorded spontaneous calcium activity in the fluorescent-identified neuron classes, and some of this activity appears coordinated between neurons within the local layer 2/3 environment. Using simultaneous stimulation by a second 2-photon laser, we correlated the connectivity of channelrhodopsin-2 expressing cells of one lineage with other neighboring neurons in layer 2/3. Connectivity based on such imaging procedures will be confirmed by optogenetics and patch-clamp electrophysiology.

Disclosures: **A. Horvath:** None. **W.A. Tyler:** None. **H. Qadir:** None. **T. Haydar:** None.

Poster

PSTR120. Neural Stem Cells and Differentiation

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR120.05/A22

Topic: A.01. Neurogenesis and Gliogenesis

Support: Collaboration for Unprecedented Success and Excellence Grant (Syracuse University; to JLM)
NIH/NINDS (JLM) 1R01NS106285]

Title: Forebrain-specific knockout of Cited2 induces transcriptome disruptions in intermediate progenitor cells, leading to altered primary cilia and disrupted neocortical development

Authors: *C. ARNELL, Jr., N. WAGNER, J. L. MACDONALD;
Syracuse Univ., Syracuse, NY

Abstract: A reduction in connectivity and synchronization between the two cerebral hemispheres is common in neurodevelopmental disorders (NDDs) including autism spectrum disorders (ASDs), intellectual disability, and schizophrenia. These interhemispheric connections are made by the axons of callosal projection neurons (CPN), a broad population of excitatory pyramidal projection neurons that are the predominant subtype of projection neuron within superficial neocortical layers (layers II/III). The transcriptional co-regulator CITED2 plays a significant role in CPN development. CITED2 is expressed by intermediate progenitor cells (IPCs) in the subventricular zone (SVZ) during the peak of layer II/III CPN generation, with a progressive refinement to CPNs of the somatosensory cortex postnatally. In *Cited2* forebrain-specific conditional knockout (cKO) mice there is a broad reduction in TBR2-positive IPCs at E15.5, leading to a reduction in the thickness of layers II/III and the neocortical length during postnatal development. Additionally, *Cited2* cKO mice exhibit reduced interhemispheric connectivity, disrupted neocortical arealization, and disorganized axonal and dendritic connectivity in the somatosensory cortex. To gain insights into the mechanisms through which CITED2 regulates IPCs, we conducted RNA-seq analysis on TBR2+ IPCs from E15.5 *Cited2* cKO mice compared to wildtype littermates. Gene Ontology analysis of the differentially expressed genes revealed upregulation of genes associated with neurons and neuronal differentiation, and downregulation of genes associated with cilia organization in the cKO IPCs. Primary cilia are essential signaling hubs that play a key role in modulating the cell cycle, in part through acting as receptors for Sonic Hedgehog signaling. We find a significant disruption in cilia length within E15.5 *Cited2* cKO IPCs compared to wildtype littermates. This disruption is consistent in CRISPR-mediated *Cited2* knockout NIH3T3 cells. These results emphasize the potential role of CITED2 in regulating primary cilia and cell cycle dynamics broadly. The *Cited2* cKO model provides a valuable system for further investigating the intricate mechanisms involved in the regulation of neuronal progenitors and neocortical development.

Disclosures: C. Arnell: None. **N. Wagner:** None. **J.L. MacDonald:** None.

Poster

PSTR120. Neural Stem Cells and Differentiation

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR120.06/A23

Topic: A.01. Neurogenesis and Gliogenesis

Support: DBT-inStem core fund
DBT/Wellcome Trust India Alliance: Intermediate Career Fellowship-IA/I/19/1/504288
DST-SERB: Start up research grant SRG/2020/001612
DBT- Har Gobind Khorana Innovative Young Biotechnologist Fellowship -BT/13/IYBA/2020/02

Pratiksha Trust grant
M.K. Bhan Young Researcher postdoctoral fellowship- HRD-12/4/2020-AFS-DBT

Title: Histone-binding protein RBBP4 regulates neurogenesis in the developing mouse neocortex

Authors: *S. K. DHANYA[1], K. KALIA, Jr.[1], T. AZAM[1], L. D'SOUZA[1], K. SWATHI[1], P. CHANDRAMOULI REDDY[2], B. MURALIDHARAN[1];
[1]Dr. Bhavana Muralidharan Lab., Inst. For Stem Cell Sci. and Regenerative Med., Bangalore, India; [2]Dept. of Life Sci., Shiv Nadar Univ., Delhi, India

Abstract: Cerebral cortex consists of diverse neuronal and glial subtypes arranged cytoarchitecturally in six layers. Dynamic interaction between chromatin modifiers drives the generation of neuronal diversity from neural progenitors. Aberrations in epigenetic modifiers result in many neurodevelopmental disorders (Stessman et al., 2017). Putative risk variants in Retinoblastoma binding protein 4 (*RBBP4*) which is a core subunit of several chromatin modifying complexes have been associated with autism spectrum disorder (ASD; Firth et al., 2009). However, the exact molecular basis by which RBBP4 functions alter the epigenetic regulation of cortical development needs to be explored. We were interested to understand the functional role of RBBP4 and to investigate its genome-wide occupancy profile in the neocortical primordium. To address this, we performed RBBP4 knockdown using the CRISPR/Cas9 approach on the neocortical progenitors at embryonic age 12.5, the stage at which deep layer neurogenesis occur producing the Layer VI and layer V neurons. Our study demonstrates that downregulation of RBBP4 in E12.5 neocortical primordium resulted in the reduction of neuronal output from progenitors, specifically decreasing the CTIP2-expressing layer V neuronal numbers with no significant impact on the TLE4-expressing layer VI neurons. Genome-wide occupancy analysis revealed that RBBP4 primarily binds to the distal regulatory elements and neuron differentiation was identified to be one of the significant GO biological pathways of RBBP4-bound genes. Interestingly, we found that RBBP4 binds to *Cdon* (Cell adhesion molecule-related/down-regulated by oncogenes), a receptor protein in the Shh signaling pathway, at the transcription start site (TSS) and distal regulatory elements. We observed that knockdown of *Cdon* resulted in a significant reduction in the generation of neurons, particularly CTIP2-positive layer V neurons. Our results shed light on the molecular and cellular role of RBBP4 and identify CDON as a novel regulator of deep layer neurogenesis in the developing neocortex.

Disclosures: S.K. Dhanya: A. Employment/Salary (full or part-time); Post doctoral fellow, Institute For Stem Cell Science and Regenerative Medicine. **K. Kalia:** A. Employment/Salary (full or part-time); Junior research Fellow, Institute For Stem Cell Science and Regenerative Medicine. **T. Azam:** A. Employment/Salary (full or part-time); Junior research Fellow, Institute For Stem Cell Science and Regenerative Medicine. **L. D'Souza:** None. **K. Swathi:** None. **P. Chandramouli Reddy:** A. Employment/Salary (full or part-time); Assistant Professor, Shiv Nadar University. **B. Muralidharan:** A. Employment/Salary (full or part-time); Assistant Professor, Institute For Stem Cell Science and Regenerative Medicine.

Poster

PSTR120. Neural Stem Cells and Differentiation

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR120.07/B1

Topic: A.01. Neurogenesis and Gliogenesis

Support: Sarah Morrison Research Award

Title: A role for Glucocorticoid Receptor Phosphorylation in Murine Stem Cell Biology and Neurogenesis

Authors: *J. TUSCHHOFF[1], N. MCCARTHY[1], P. MONAGHAN-NICHOLS[2];
[1]Univ. of Missouri, Kansas City, Kansas City, MO; [2]Dept. of Basic Med. Sci., UMKC Sch. of Med., Kansas City, MO

Abstract: Background: Synthetic glucocorticoids (sGCs) are administered to women at risk for preterm labor to promote lung maturation and to reduce premature birth risks to the child. Animal studies indicate that prenatal exposure to sGC induced changes in neural stem cell fate, and neurological and behavioral changes. Outcomes sometimes associated with exposure to sGCs during development in humans include depression, Attention Deficit Hyperactivity Disorder, and language problems associated with a higher basal hypothalamic-pituitary axis (HPA) activity and higher levels of anxiety and depression. In studies on the relationship between leukocyte activation and psychiatric disorders, differential ratios of phosphorylation on serine 211 and serine 226 (S211 and S226) on the glucocorticoid receptor (GR) were associated with increased risks for mood disorders. Objectives: To assess the role of glucocorticoid receptor phosphorylation on cell fate and neurogenesis in the developing brain. Approach: Mice were genetically engineered with to replace the murine equivalents of human serine 211 (mouse S220) and human serine 226 equivalent (mouse S234) with alanine (S220A:S234A). Neural stem cell cultures were established from cerebral cortices in control and experimental animals, and exposed to different sGC and their genomic and biological consequences examined using whole genome expression studies, cell proliferation and cell fate assays in-vitro. Knock in and wild type embryos were treated with sGC or vehicle on embryonic day 14 in -vivo, and the anatomical consequences examined in basal and stimulated conditions compared to control using immunohistochemical staining for cell type specific markers. Results: Genome studies indicate that basal phosphorylation on S220 and S234 is required to maintain stem cell function and that loss of phosphorylation on S220 and S234 activate distinct transcriptomes basally and in response to sGC stimulation. In-vivo studies indicate that phosphorylation on S211 and S226 is required for cell fate specification and anatomical organization of the developing cerebral cortex.

Disclosures: J. Tuschhoff: None. **N. McCarthy:** None. **P. Monaghan-Nichols:** A. Employment/Salary (full or part-time); UMKC School of Medicine.

Poster

PSTR120. Neural Stem Cells and Differentiation

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR120.08/B2

Topic: A.01. Neurogenesis and Gliogenesis

Support: NIH/NINDS Grant#R01NS111997

Title: Determining the proteome of in vivo mouse cortical neuronal differentiation

Authors: *V. DAL POZZO[1], C. SMITH[1], Y. XIE[2], Z. LIN[2], P. SURANA[3], F. VITORINO[2], R. DAVULURI[3], B. GARCIA[2], N. DAHMANE[1];
[1]Cornell University: Weill Cornell Med. Col., New York, NY; [2]Dept. of Biochem. and Mol. Biophysics, Washington Univ. Sch. of Med., St. Louis, MO; [3]Dept. of Biomed. Informatics, Stony Brook Cancer Ctr., Stony Brook, NY

Abstract: The development of the mouse cerebral neocortex is a complex and closely regulated sequence of neuronal production, migration, and differentiation. During cortical neurogenesis from Embryonic day (E)10.5 to about E18.5, two major types of precursors have been described: multipotent radial glial stem/progenitor cells (RGCs) in the ventricular zone (VZ) and derived intermediate neurogenic progenitors (INPs) or basal progenitors, in the subventricular zone (SVZ). Both RGCs and INPs can generate neurons that will constitute the 6-layered mammalian cerebral cortex. We have devised a sorting procedure to isolate these cell types from the embryonic neocortex (Xiang et al, *Cell Death and Differentiation* 2012) that allow us to purify from embryonic cortices RGCs, INPs and postmitotic neurons. We have used RGCs, INPs and postmitotic neuronal cells purified from E14.5 mouse cortices to characterize the whole cell proteome of the *in vivo* embryonic cortical cells using liquid chromatography-tandem mass spectrometry (LC-MS/MS). We analyzed three replicates of each sorted cell type, combined analysis of the replicates using Proteome Discoverer (v3.0) software and applied a peptide and protein false discovery rate (FDR) of 1% identified more than 5600 proteins. Notably, the expression of several proteins was consistent with the previous observation using immunofluorescence, such as PAX6 (paired box protein Pax-6), TBR2 or EOMES (eomesodermin homolog), and TBR1 (T-box brain protein 1) which are sequentially enriched by RGCs, INPs, and postmitotic neurons, respectively. Furthermore, we grouped the proteins based on their expression level and analyzed their biological functions based on the gene ontology (GO) and observed a regulation of proteins involved in mRNA processing during neuronal differentiation. We also compared our proteomic data to proteomic data obtained on *in vitro* differentiation systems as well as to single cell RNA-seq datasets obtained from mouse embryonic cortex to correlate cortical cells proteome and transcriptome. While many studies have investigated the proteome of cells undergoing neuronal differentiation *in vitro*, ours has used quantitative proteomics to decipher the proteome of *in vivo* embryonic cortical cell types to identify novel regulators of mammalian neuronal differentiation.

Disclosures: V. Dal Pozzo: None. **C. Smith:** None. **Y. Xie:** None. **Z. Lin:** None. **P. Surana:** None. **F. Vitorino:** None. **R. Davuluri:** None. **B. Garcia:** None. **N. Dahmane:** None.

Poster

PSTR120. Neural Stem Cells and Differentiation

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR120.09/B3

Topic: A.01. Neurogenesis and Gliogenesis

Title: Rbfox2 regulates neuronal differentiation and migration in the developing cortex via time-specific alternative splicing.

Authors: *S. WEIßBACH[1], *S. WEIßBACH[3], H. TODOROV[4], M. HEINE[2], S. GERBER[4], J. WINTER[5];
[1]nstitute of Developmental Biol. and Neurobio. (iDN), [2]Inst. of Developmental Biol. and Neurobio. (iDN), Johannes Gutenberg Univ., Mainz, Germany; [3]Inst. of Developmental Biol. and Neurobio. (iDN), Univ. Mainz, Mainz, Germany; [4]Inst. of Human Genet., [5]Inst. for Human Genet., Univ. Med. Ctr. of the Johannes Gutenberg Univ., Mainz, Germany

Abstract: Neurogenesis is a complex process orchestrated by a myriad of intricate biological mechanisms, among which alternative splicing has emerged as a fundamental regulatory mechanism. Alternative splicing enables the expression of multiple isoforms from a single gene. Indeed, it plays a crucial role in the transition from neuronal progenitor cells to neurons and additionally shapes the functional diversity of neuronal cells during development. This process allows for precise control over protein function, localization, and interactions, thereby influencing critical neuronal differentiation and migration aspects. The RBFOX family of proteins has gained considerable attention due to their indispensable role in brain development. In particular, *Rbfox2* has been shown to be highly abundant in the neocortex prenatally at the peak of neurogenesis. At this time of development, RBFOX2 is only weakly expressed in neural stem and progenitor cells and highly expressed in differentiating newborn neurons suggesting an important role during neural differentiation. Here, we aimed at elucidating the role of RBFOX2-mediated alternative splicing in neuronal cell differentiation and migration. Using the in-utero electroporation technique, we induced *Rbfox2* overexpression at E13 and conducted a comprehensive molecular characterization of the RBFOX2 overexpressing neocortex at E15. Our results revealed a distinct cellular phenotype associated with *Rbfox2* overexpression. Specifically, we observed a significantly reduced differentiation of neural progenitor cells into mature neurons. To investigate the underlying basis of this phenotype, we performed RNA sequencing and identified numerous developmentally relevant alternative splicing targets directly regulated by RBFOX2. Additionally, we discovered an antagonistic relationship between RBFOX2 and PTBP1 in the regulation of specific exons. Taken together our data indicates that RBFOX2 shapes neuronal differentiation through time-specific alternative splicing.

Disclosures: S. Weißbach: None. **S. Weißbach:** None. **H. Todorov:** None. **M. Heine:** None. **S. Gerber:** None. **J. Winter:** None.

Poster

PSTR120. Neural Stem Cells and Differentiation

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR120.10/B4

Topic: A.01. Neurogenesis and Gliogenesis

Title: Mechanisms of early development of the cerebral cortex in models of 16p11.2 genetic locus Copy Number Variations

Authors: O. SQUIRE, C. ZUGLIAN, C. WOOD, I. MORELLA, J. HALL, R. BRAMBILLA, ***F. BEDOGNI**;
Cardiff Univ., Cardiff, United Kingdom

Abstract: Excitatory neurons of the cerebral cortex are generated during embryonic life, when proliferating cortical progenitors either regenerate themselves and re-enter the cell cycle or exit the cell cycle to differentiate into postmitotic cells. The importance of such early events is well illustrated by the fact that transition through the cell cycle is often affected in multiple neurodevelopmental disorders (NDDs). Accordingly, symptoms associated with NDDs often overlap, and typically arise from impaired neuronal differentiation and poor maturity of neuronal networks in the cerebral cortex. This suggests that common, core developmental processes may be disrupted across a range of NDDs. G1 phase of the cell cycle in particular plays an important role in neuronal differentiation. G1 phase lengthens across cortical neurogenesis, with inner cortical layer neurons generated first through a relatively short G1 transition, and outer cortical layer neurons, generated later, arising from longer G1 transitions. It is known that changes in the duration of G1 impair differentiation in multiple NDD models, and we show that this is accompanied by changes in the levels of Neurod1. Neurod1 is a pro-neurogenic transcription factor involved in chromatin changes that is expressed by cortical progenitors transitioning from G1 phase to postmitotic differentiation. Accordingly, aberrant cell cycle transition and deregulated levels of Neurod1 are displayed in models of Copy Number Variations (CNVs) of the 16p11.2 genomic locus, a high risk factor for NDDs. Although 16p11.2 duplication carriers bear greater cognitive impairments and increased risk of psychosis compared to deletion carriers, people harboring locus deletion or duplication both display intellectual disability and autistic traits. This suggests that symptoms associated with 16p11.2 CNVs are largely overlapping. In line with this, we show that both deletion and duplication of the 16p11.2 locus reduces the number of basal progenitors. Considering the key role of basal progenitors in cortical development, our data suggest that similar differentiation impairments may arise from seemingly opposite regulation of the timing of proliferation driven by either deletion or duplication of the 16p11.2 locus. Cumulatively, this evidence further supports the existence of common pathogenic mechanisms at the root of multiple NDDs.

Disclosures: O. Squire: None. **C. Zuglian:** None. **C. Wood:** None. **I. Morella:** None. **J. Hall:** None. **R. Brambilla:** None. **F. Bedogni:** None.

Poster

PSTR120. Neural Stem Cells and Differentiation

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR120.11/B5

Topic: A.01. Neurogenesis and Gliogenesis

Support: NICHD Intramural Support

Title: Differential activity of the transcription factor Nkx2.1 in embryonic mouse brain, lung, and thyroid

Authors: *M. T. C. MANION[1,2], M. SOHN[3], Y. ZHANG[2], P. ROCHA[4], R. K. DALE[3], T. J. PETROS[2];
[1]NICHD, Bethesda, MD; [2]Unit on Cell. and Mol. Neurodevelopment, Eunice Kennedy Shriver Natl. Inst. of Child Hlth. and Human Develop. (NICHD), Bethesda, MD; [3]Bioinformatics and Scientific Programming Core, Eunice Kennedy Shriver Natl. Inst. of Child Hlth. and Human Develop. (NICHD), NIH, Bethesda, MD 20892, USA, Bethesda, MD; [4]Unit on Genome Structure and Regulation, Eunice Kennedy Shriver Natl. Inst. of Child Hlth. and Human Develop. (NICHD), NIH, Bethesda, MD 20892, USA, Bethesda, MD

Abstract: Neural inhibition, which is critical for proper brain function, is dependent on GABAergic interneurons, a heterogeneous cell population with dozens of subtypes displaying distinct morphologies, connectivity, electrophysiology properties, neurochemical markers, and gene expression. A major population of GABAergic interneurons arises from the media ganglionic eminence (MGE), one of several transient developmental structures in the embryonic brain. The transcription factor Nkx2.1 (previously thyroid transcription factor 1 - Ttf-1 - reflecting its original discovery) is a master regulator required for all MGE-derived GABAergic neurons. Nkx2.1 is also instrumental in for proper development of lung and thyroid tissue, and humans with *NKX2.1* mutations suffer from Brain-Lung-Thyroid Syndrome. While the function of Nkx2.1 has been studied separately in the MGE, lung, and thyroid, the full picture of how Nkx2.1 interactions regulate distinct transcriptional programs in each of these regions is not known. To probe differential Nkx2.1 binding locations in MGE, lung, and thyroid, we have collected tissue from MGE, lung, and thyroid at embryonic day 13.5 (E13.5) and used CUT&RUN, a variant on ChIP-seq, to identify DNA regions bound by Nkx2.1. These results reveal both global Nkx2.1 interaction sites in all three tissues, as well as organ-specific interactions at specific loci. We also performed single cell Multiome sequencing (snATAC-seq and snRNA-seq) to analyze chromatin accessibility and gene expression in these 3 different tissues. In sum, we have characterized how Nkx2.1 regulates the transcriptional network and cell fate in the embryonic MGE, lung and thyroid.

Disclosures: M.T.C. Manion: None. **M. Sohn:** None. **Y. Zhang:** None. **P. Rocha:** None. **R.K. Dale:** None. **T.J. Petros:** None.

Poster

PSTR120. Neural Stem Cells and Differentiation

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR120.12/B6

Topic: A.01. Neurogenesis and Gliogenesis

Support: ERC Grant EC787355

Title: Differential deployment and programmed cell death shape region specific interneuron cytoarchitectures in the cerebral cortex

Authors: *E. J. PAUL[1], E. SERAFEIMIDOU POULIOU[2], O. MARIN[3];
[1]Ctr. for Developmental Neurobio., Kings Col. London, London, United Kingdom; [2]Ctr. for Developmental Neurosci., [3]King's Col. London, London, United Kingdom

Abstract: Consisting of many functionally distinct areas the cortex is one of the most complex regions of the mammalian brain. Understanding how this diversification occurs during development remains a major goal of neuroscience. Interneurons in the adult mouse cortex are a hugely diverse cell population that are broadly grouped based on developmental origin (MGE or CGE) and can be defined by distinct morphology, physiological properties, and expression of molecular markers (e.g., parvalbumin+ basket cells or VIP+ bipolar cells). Despite making up approximately one sixth of the total neuronal population a cortex-wide map of GABA populations is currently missing from the literature.Using a combination of Cre/flp-driver mouse lines crossed with fluorescent reporter lines we have mapped the overall distribution of interneurons, of broad populations (MGE and CGE derived) and of five discreet populations (PV-Basket, T-shaped martinotti, fanning out martinotti, VIP-bipolar and CCK-basket) across 26 distinct cortical regions in the adult mouse. We have shown that interneuron distribution is extremely heterogenous across the cortex. We demonstrate this using cell density, ratio to pyramidal cells and the proportion of total interneuron population. Our data suggests specific inhibitory motifs may be present at varying levels across cortical regions.The final number of cells in the cortex is fine-tuned via programmed cell death, which occurs during early postnatal development, and is important for maintaining the correct balance of excitatory and inhibitory cells. This normal developmental process has been studied in the mouse somatosensory cortex but has not yet been examined across the entire cortex. We used transgenic knock-out mice in which GABA cell populations are ‘immortalised’ to prevent programmed cell death. We then compared densities to those in control tissue to determine the degree of cell death and create an atlas of programmed cell death in the cortex. We found that levels of cell death of interneurons across the mouse cortex is heterogeneous and ranges from 0-40%. A degree of heterogeneity is still present across the cortex when interneurons are immortalised suggesting that there is also differential deployment during development. The atlases we have developed will be made available online and will be helpful to those studying the cortex, a region that should not be assumed uniform in the distribution of inhibitory cell populations. Further investigation could establish whether these differences in inhibitory populations and the prevalence of different inhibitory motifs contributes to the functional specialisation of cortical areas.

Disclosures: **E.J. Paul:** None. **E. Serafeimidou Pouliou:** None. **O. Marin:** None.

Poster

PSTR120. Neural Stem Cells and Differentiation

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR120.13/B7

Topic: A.01. Neurogenesis and Gliogenesis

Support: NICHD Intramural support

Title: Histone 3.3 lysine 4 methionine mutation of medial ganglionic eminence-derived interneuron causes developmental defects

Authors: *J. LI, D. ABEBE, M. MANION, S. BEYENE, D. LEE, Y. ZHANG, T. PETROS; NICHD/Eunice Kennedy Shriver Natl. Inst. of Child Hlth. and Human Develop., Bethesda, MD

Abstract: Methylation of lysine 4 on histone H3 (H3K4) is required for promoter activation and undergoes dynamic regulation at gene loci during neurodevelopment. Perturbation in H3K4 methylation is associated with several neurodevelopmental syndromes, but the specific role of H3K4 methylation in interneuron development has not been characterized. Here, we explore the role of H3K4 methylation in medial ganglionic eminence (MGE)-derived interneurons by directly mutating the lysine 4 on H3.3 to the methionine (H3.3K4M). We found that the expression of H3K4 methylation in MGE is decreased at E13.5. Compared with control mice, H3.3K4M mutant mice showed significant growth retardation and increased death during the 2nd-3rd postnatal weeks. Despite the mutant mice being significantly smaller than control mice early on, they became obese during adolescence. Mutation of H3.3K4M decreased the number of MGE-derived cortical interneurons at P21, specifically a reduction in the number of PV+ and SST+ interneurons in the neocortex. We also found that the mutant mice showed impaired locomotor activity and exploration behavior by home-cages test and open field test. In summary, our results reveal that perturbation of H3K4 methylation alters interneuron fate and causes profound developmental and behavioral defects.

Disclosures: J. Li: None. **D. Abebe:** None. **M. Manion:** None. **S. Beyene:** None. **D. Lee:** None. **Y. Zhang:** None. **T. Petros:** None.

Poster

PSTR120. Neural Stem Cells and Differentiation

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR120.14/B8

Topic: A.01. Neurogenesis and Gliogenesis

Support: CDMRP Grant W81XWH-21-1-0585

Title: Ras/mapk signaling impacts in gabaergic cortical interneuron development

Authors: A. M. STAFFORD[1], S. J. KNOWLES[2], D. PACHECO-CRUZ[1], J. M. NEWBERN[2], ***D. VOGT**[1];
[1]Michigan State Univ., Grand Rapids, MI; [2]Arizona State Univ., Tempe, AZ

Abstract: The RAS/MAPK pathway is a highly conserved signaling cascade found in nearly all cells. Multiple genes are involved in this pathway, resulting in distinct syndromes with unique and overlapping cognitive symptoms. The molecular and cellular mechanisms that lead to these symptoms are still being investigated. One group of cells that may underlie some of these symptoms are GABAergic cortical interneurons (CINs). We and others previously showed that the proportion of parvalbumin and somatostatin expressing CINs is shifted in multiple RAS/MAPK genetic mouse mutants and some core GABAergic developmental programs are regulated by RAS/MAPK signaling, including some cardinal transcription factors. Here, we expand upon these initial findings to explore behavioral consequences relevant to the associated syndromes and additional cellular and molecular phenotypes during development. In addition, we are testing whether an FDA-approved drug that inhibits RAS/MAPK signaling may alleviate some of the phenotypes and show promise towards those impacted by the various RAS/MAPK syndromes.

Disclosures: A.M. Stafford: None. **S.J. Knowles:** None. **D. Pacheco-Cruz:** None. **J.M. Newbern:** None. **D. Vogt:** None.

Poster

PSTR120. Neural Stem Cells and Differentiation

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR120.15/B9

Topic: A.01. Neurogenesis and Gliogenesis

Support: NIMH Grant R00MH126430-03

Title: Molecular Mechanism of Ryk Regulating Interneuron Differentiation in Medial Ganglionic Eminence

Authors: *Y. CAO, R. LIAQAT, M. CAMPBELL;
UCSD Dept. of Neurosciences, La Jolla, CA

Abstract: Interneurons play critical roles in modulating cortical circuitry and plasticity. There are many subtypes of interneurons with relatively uniform neurochemical, morphological, and electrophysiological properties in the neocortex. Somatostatin- (SST+) and parvalbumin-positive (PV+) interneurons are two major subtypes which are generated from medial ganglionic eminence (MGE) during embryonic stages. It is known that the neurogenesis of SST+ and PV+

interneurons in MGE is regulated both spatially and temporally. However, it is still unclear how progenitors in different regions of MGE give rise to different subtypes of interneurons. Our recent study shows that Ryk is highly involved in cell-fate determination in MGE along the rostral-caudal axis (McKenzie et.al. 2009). Caudal MGE, with higher levels of nuclear Ryk, generates more SST+ interneurons, and rostral MGE, with lower Ryk, gives rise to more PV+ interneurons. Ryk is a kinase-dead thymidine kinase-like receptor that binds and responds to Wnt. It's known that Ryk doesn't have any DNA binding domains to directly modulate gene expression. The molecular mechanism of Ryk's function during interneuron differentiation in MGE is largely unclear. To further investigate this, we applied a biotin-streptavidin pull-down and mass-spectrometry analysis to find the potential binding partners of Ryk. Next, we test if the inhibition of these candidates could bias the SST+/PV+ ratio in mouse embryonic stem cells (ESCs) induced neurons and in vivo by transplanting these induced neurons. Taking advantage of bulk and single-cell RNA sequencing, we identified downstream regulatory modules of Ryk and its binding partners. Our findings further elucidate the spatial and temporal mechanisms that regulate cell-fate determination in MGE.

Disclosures: **Y. Cao:** None. **R. Liaqat:** None. **M. Campbell:** None.

Poster

PSTR120. Neural Stem Cells and Differentiation

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR120.16/B10

Topic: A.01. Neurogenesis and Gliogenesis

Support: NSFC Grant 32100786

Title: Epigenetic factors regulate cortical interneuron development

Authors: *T. TANG[1], Y. XIE[2];
[2]Fudan Univ., [1]Fudan Univ., Shanghai, China

Abstract: Objective The balance of excitation and inhibition in cortical circuits is depended on the appropriate number of pyramidal neurons and inhibitory neurons which are derived from different regional neural stem cells. Our previous results show that cortical neurogenesis depends on the positioning of intermediate progenitors. This process is safeguarded by epigenetic factors widely expressed in progenitors and neurons. However, whether these epigenetic factors regulate the neurogenesis of cortical interneurons remains unclearly. Methods The target genes were conditionally knockout in cortical interneurons from neural stem cells with the Cre-LoxP system. Immunofluorescence staining and multiple next-generation sequencing were performed to analyze the phenotype and the mechanism. Results (1) The number and migration of cortical interneurons were affected in the mutant mice. (2) The cell fate determination of interneuron progenitors was changed. (3) The cell-fate changed neurons showed abnormal decision of positioning. Conclusion The epigenetic factors regulate the neurogenesis of cortical interneurons.

Disclosures: **T. Tang:** None. **Y. Xie:** None.

Poster

PSTR120. Neural Stem Cells and Differentiation

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR120.17/B11

Topic: A.01. Neurogenesis and Gliogenesis

Support: NICHD Intramural Support

Title: A spatial organization within the caudal ganglionic eminence relating to mature interneuron subtypes

Authors: *S. BEYENE, M. T. MANION, Y. ZHANG, T. J. PETROS;
Eunice Kennedy Shriver Natl. Inst. of Child Hlth. an, Bethesda, MD

Abstract: One of the fundamental questions of developmental neuroscience is how do neurons acquire specialized molecular, morphological, and electrophysiological fates in the mammalian forebrain? This question is pertinent to the study of cortical inhibitory interneurons (cINs), an extremely heterogeneous group of GABAergic cells whose dysfunction is related to numerous neurodevelopmental disorders such as epilepsy, schizophrenia and autism. Most of these GABAergic interneurons arise from two germinal zones of the developing brain termed the medial and caudal ganglionic eminences (MGE and CGE, respectively). The relationship between spatial subdomains within the MGE and mature interneuron subtype was revealed over 15 years ago using microdissection and transplantation techniques. In the MGE, somatostatin (SST) cINs preferentially arise from the dorsal MGE whereas parvalbumin (PV) cINs arise from the ventral MGE. However, a similar relationship has not been explored in the CGE. Here, we characterize the spatial origin of distinct CGE-derived interneuron subtypes by harvesting anterior and posterior CGE tissue at 2 different timepoints (E13.5 and E15.5) from Dlx5/6-Cre;Ai9 mice and transplanted these cells into the cortex of neonatal WT hosts (P0-P4). Immunostaining brains for CGE-derived interneuron markers 30 days post transplantation (P35) has revealed Vasoactive Intestinal Peptide (VIP)- and Cholecystokinin (CCK)-expressing cINs preferentially derived from the anterior CGE, whereas there is a bias for Reelin (Rln)-expressing cINs originating from the posterior CGE. Linking this knowledge to spatial transcriptomics data will advance our understanding of how CGE microdomains regulate cIN fate decisions during normal development and possibly in neurological disorders.

Disclosures: **S. Beyene:** None. **M.T. Manion:** None. **Y. Zhang:** None. **T.J. Petros:** None.

Poster

PSTR120. Neural Stem Cells and Differentiation

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR120.18/B12

Topic: A.08. Development of Neural Systems

Support: NIH R01 NS093009 to V.V.C

Title: Identification of genes misregulated in the cortical hem of dreher mice.

Authors: *I. ISKUSNYKH[1], N. FATTAKHOV[1], M. KIRCHNER[1], A. ZAKHAROVA[1], L. MUKHAMETZYANOVA[1], Y. LI[2], L. BIHANNIC[2], E. STESHINA[1], P. NORTHCOTT[2], V. CHIZHIKOV[1];
[1]Univ. of Tennessee Hlth. Sci. Ctr., Memphis, TN; [2]St Jude Children's Res. Hosp., Memphis, TN

Abstract: The cortical hem (CH) is a transient neuroepithelial structure that gives rise to the Cajal-Retzius cells (CR). It is located between the choroid plexus and hippocampus in the developing telencephalon. As part of the hippocampus where adult neurogenesis occurs, the dentate gyrus (DG) plays an important role regulating spatial behavior and memory formation. Depression, psychosis, and addiction are some examples of psychiatric disorders which may arise due to DG dysfunction. In our previous studies, we found that *Lmx1a*-deficient (*dreher*) mice had a compromised transhilar radial glial scaffold, reduced proliferation and neurogenesis in the hippocampal primordium, and impaired radial migration of DG progenitors. However, the mechanisms by which *Lmx1a* regulates DG development remain largely unknown. Using RNAseq analysis of CH samples obtained by laser capture microdissection from wild type and *dreher* embryos, we have found that loss of *Lmx1a* downregulates 612 and upregulates 457 genes in the CH. Our gene ontology analysis revealed that misregulated genes belong to various categories, such as transcription factors (Lmo2), transmembrane receptors (Plxnc1), lipid and carbohydrate metabolism regulators (Ccdc3, Rhd10), and transporters of molecules (Slc17a8, Slc14a2). We have identified 13 novel CH markers (*Slc17a8, Ccdc3, Adamts1, Galnt14, Camk2a, Arfgef3, Rdh10, Plxnc1, Kdr, Asb4, Slc14a2, Peg3, Slit3*) downregulated in *dreher* embryos. Interestingly, loss of *Lmx1a* also leads to downregulation of CR cell-enriched genes (*Car10, Lhx1, Trp73, Cntnap2, Reln, Cdkn1a, Chst8, Spock1, Akap6, Cacna2d2, Tbr2*). Thus, the ablation of *Lmx1a* affects the expression of CH markers and compromises the expression of genes associated with CR development.

Disclosures: I. Iskusnykh: None. **N. Fattakhov:** None. **M. Kirchner:** None. **A. Zakharova:** None. **L. Mukhametzyanova:** None. **Y. Li:** None. **L. Bihannic:** None. **E. Steshina:** None. **P. Northcott:** None. **V. Chizhikov:** None.

Poster

PSTR120. Neural Stem Cells and Differentiation

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR120.19/B13

Topic: A.01. Neurogenesis and Gliogenesis

Support: Tashia and John Morgridge Endowed Faculty Scholar in Pediatric Translational Medicine of the Stanford Maternal and Child Health Research Institute
Brain and Behavior Foundation (NARSAD young investigator award)
Carol Lavin Bernick Faculty Grant Program

Title: Micrornas in the 12qf1 cluster influence corticothalamic projection neuron fate by regulating expression of the transcriptional co-repressor tle4

Authors: *S. THARIN[1], V. SITHTHANANDAN[1], M. GALAZO[2];
[1]Stanford Univ., Palo Alto, CA; [2]Tulane Univ., New Orleans, LA

Abstract: Corticofugal projection neurons (CFuPN) are one of the defining features of the mammalian brain. They have a unique role in transmitting signals from the cerebral cortex to lower brain regions and the spinal cord. Subcerebral projection neurons (SCPN) and corticothalamic projection neurons (CThPN) are two closely related CFuPN subtypes. In mice, CThPN reach peak production around e12.5 and populate layer VI of the cortex. These neurons, which project to the thalamus and modulate its activity, have an important role in sensory and cognitive function. SCPN, including corticospinal motor neurons (CSMN) which form the corticospinal tract and are involved in motor function, reach peak production around e13.5 and populate layer V. In our previous work we identified microRNAs (miRs) that are selectively expressed by mouse CSMN verses callosal projection neurons (CPN) during development. A detailed study of one of those miRs, miR-409-3p, showed that it represses LMO4, a transcriptional activator expressed by CPN, and can control corticospinal fate. Additionally, many of the miRs enriched in CSMN verses CPN, including miR-409-3p, were found to be encoded in a large miR cluster at 12qF1. This enrichment, coupled with varying levels of expression of multiple genes associated with both CSMN and CThPN cell fate, led us to ask: are any miRs of the cluster also able to influence CSMN verses CThPN fate by regulation of those genes? Here we show that two other 12qF1-encoded miRs - miR-369-3p and miR-496a-3p - repress TLE4, a transcriptional co-repressor associated with development of CThPN identity. Luciferase reporter gene assays confirmed a direct interaction between both miR-369-3p and miR-496a-3p with their respective predicted binding sites in the TLE4 3’ untranslated region. Overexpression of miR-496-3p in primary cultures of embryonic cortical neurons results in a decrease in the percentage of CThPN, relative to scrambled control, as would be predicted if miR-496a-3p were repressing TLE4 in cortical progenitors. Our findings demonstrate another example of the important role miRs, and specifically those of the cluster found at 12qF1, have in selectively repressing transcriptional regulators within related neuronal subtypes, thereby ensuring proper cortical development.

Disclosures: S. Tharin: None. **V. Siththanandan:** None. **M. Galazo:** None.

Poster

PSTR120. Neural Stem Cells and Differentiation

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR120.20/B14

Topic: A.01. Neurogenesis and Gliogenesis

Support: NIH Grant MH113257 (to AD)

Title: Relationship between interstitial neurons of the cerebral white matter and layer 5 cortical neurons in the macaque: clinical implication

Authors: *B. AHMED[1], A. DUQUE[2], P. RAKIC[3], Z. MOLNAR[4];
[1]DPAG, Sherrington Building, Univ. of Oxford, Oxford, United Kingdom; [2]Yale Univ. Sch. Med., Yale Univ. Sch. Med., New Haven, CT; [3]Yale Univ. Sch. Med, Dept of Neurosci., Yale Univ. Sch. Med, Dept of Neurosci., New Haven, CT; [4]Univ. Oxford, Univ. Oxford, Oxford, United Kingdom

Abstract: Interstitial neurons of the cerebral white matter (WMICs) are heterogeneously distributed within the entire subcortical white matter and in number form approximately 3% of all cortical neurons[1]. WMICs are known to be the remnants of neurons found within the subplate and during development undergo cell death such that in the adult only 20% survive. WMICs form a diverse population both neurochemically and morphologically[2]. They have been implicated in pathology, especially schizophrenia and epilepsy, where an increased density of interstitial neurons has been observed[3,4].
We have examined high quality images of histological sections of NeuN immuno-labeled neurons of the Macaque cortex available in Collection 6 of MacBrainResource located in the Department of Neuroscience at Yale University School of Medicine (https://medicine.yale.edu/neuroscience/macbrain/). The distribution of labelled neurons in the images were detected by the image analysis software QuPath (Bankhead, P. et al. (2017). QuPath: Open source software for digital pathology image analysis**.** *Scientific Reports* 7, 16878). We have analysed coronal sections, 1 cm apart, from anterior to posterior of the right hemisphere, and quantified neuronal numbers in Layers 5, 6a, 6b and white matter in three regions of the cortex: Linear (unfolded cortex with short parallel laminae), Sulcal and Gyral. We confirm that the densities of WMICs are highest in Gyral crowns, intermediate in Linear regions, and least at Sulci. We also confirm that the thickness of Layers 5, 6a, and 6b are thickest at Gyral crowns, intermediate in Linear regions, and thinnest at Sulci. By optimising QuPath parameters, we have quantified the number of NeuN labelled neurons in Layers 5, 6a, 6b, and interstitial cells underlying Layer 6b-white matter boundary at Gyri, Linear and Sulci regions. Based on a normalization protocol, our quantitative analyses have shown a consistent relationship between the number of WMICs and the number of Layer 5 cortical neurons in these three regions. We find that for each interstitial cell there are of the order of 5 cortical neurons of Layer 5, irrespective of whether the regions are located at Gyri, Linear or Sulci.
We propose that the survival of WMICs is dependent upon the number of Layer 5 cortical neurons. Furthermore, in pathological conditions, we propose the change in density or distribution of WMICs is a consequence *per se* of modification in the number of Layer 5 neurons.
1. Sedmak G and Judas M (2019). J Anat. 235, 626-636.
2. Kostovic I and Rakic P (1980). J Neurocytol. 9, 219-242.

3. Anderson SA, Volk DW, and Lewis DA (1996). Schizophr Res. 19, 111-119.
4. Meencke HJ (1983). J Neurol. 230, 171-181.

Disclosures: **B. Ahmed:** None. **A. Duque:** None. **P. Rakic:** None. **Z. Molnar:** None.

Poster

PSTR121. Autism: Behavioral and Neural Mechanisms

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR121.01/B15

Topic: A.07. Developmental Disorders

Title: Developmental PFOA Exposure Increases Neural Activation in Sensory Neurons and Induces Behavioral Pathology Relevant to Autism Spectrum Disorders

Authors: *J. J. MERLINO[1,2], T. R. GORUK[1], M. SCOTTO[1], R. ST. LAURENT[1], T. OLIVA[2], N. MORMILE[2], K. DIGIULIO[2], M. LAURIA[2], C. BARBARO[2], S. GUARIGLIA[1,2];
[1]Developmental Neurobio., Inst. for Basic Res. in Developmental Disabilities, Staten Island, NY; [2]St. Joseph by the Sea High Sch., Staten Island, NY

Abstract: Perfluorooctanoic acid (PFOA) is a widely present environmental contaminant found in various consumer products and food sources. It has been linked to negative health consequences, some of which include disrupted reproductive, endocrine, and immune system function, as well as an increased risk of cancer. In this study, we investigated the impact of early developmental exposure to PFOA on behaviors related to autism spectrum disorders (ASD) using a species of planarian called *Dugesia dorotocephalata*. We focused on examining locomotor activity, exploratory behavior, social aggregation, and sensory neuron activation in the planarians. The animals were exposed to three different concentrations of PFOA (20, 200, and 2000 PPT) immediately after decapitation and throughout the initial seven days of neurodevelopment, which allows for full brain regeneration. To assess planarian activity, we utilized a 5 cm concave open field and AnyMaze software, which provided a high contrast between the animals and the arena background, enabling robust movement analysis. Interestingly, the total distance covered in the open field did not differ significantly between the experimental groups ($F_{(3.00, 58.04)} = 1.053$; $p = 0.3561$; $n = 79$). However, both the time spent in the center of the open field ($F_{(3.00, 64.34)} = 6.743$; $p = 0.005$; $n = 79$) and the distance traveled in the center ($F_{(3.00, 73.98)} = 5.680$; $p = 0.0015$; $n = 79$) were significantly greater in the 20 and 200 PPT groups compared to the control group. These groups also exhibited repetitive turning on their center of mass ($F_{(3.00, 68.22)} = 4.903$; $p = 0.0031$; $n = 79$), indicating a hyperactive phenotype possibly driven by stress. Furthermore, planarians exposed to PFOA displayed increased c-Fos expression in ganglion and sensory neurons of the head, suggesting heightened sensory sensitivity to the novel planarian water encountered in the experiment. This heightened sensory perception is commonly observed in individuals with ASD. Importantly, developmental exposure to PFOA led to a reduction in social clustering behavior among the planarians ($F_{(3, 172)} = 3.037$; $p = 0.0031$, $n = 176$), which may also be attributed to heightened sensory sensitivity. In summary,

our findings demonstrate that PFOA exposure during early development induces increased sensitivity to chemical cues from other planarians, hyperactivity, atypical exploration of novel environments, including repetitive turning, and impaired social clustering. We propose that the altered sensory processing resulting from PFOA exposure contributes to these behavioral changes, which closely resemble behaviors observed in individuals with ASD.

Disclosures: J.J. Merlino: None. **T.R. Goruk:** None. **M. Scotto:** None. **R. St. Laurent:** None. **T. Oliva:** None. **N. Mormile:** None. **K. DiGiulio:** None. **M. Lauria:** None. **C. Barbaro:** None. **S. Guariglia:** None.

Poster

PSTR121. Autism: Behavioral and Neural Mechanisms

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR121.02/B16

Topic: A.07. Developmental Disorders

Title: Gut Microbiota Disruption and Planarian Social Behavior: Unveiling the Impact of Antibiotics and Bifidobacterium Probiotics

Authors: *M. DODGE[1], M. DANIELS[2], A. REYNOLDS[2], M. SCOTTO[1], J. LICHAA[1], S. GUARIGLIA[1,3];
[1]New York State Inst. for Basic Res., Staten Island, NY; [2]Wagner Col., Staten Island, NY; [3]St. Joseph by the Sea High Sch., Staten Island, NY

Abstract: The gut is inhabited by a vast and diverse community of microorganisms, including bacteria, archaea, and eukaryotes, with an estimated human-to-bacterial cell ratio of 1:1. The gastrointestinal tract is home to over 10^14 microorganisms. Probiotics, defined as live microorganisms that provide health benefits when administered in sufficient quantities, have gained recognition. Bifidobacterium longum, a commonly used probiotic strain, has demonstrated numerous positive effects on human health, including anti-infection activity, regulation of psychological well-being, and enhancement of host nutrient absorption. However, the effects of probiotics, specifically Bifidobacterium, on planarian behavior remain largely unexplored. This work investigated the impact of Bifidobacterium on planarian social behavior. The hypothesis proposes that treating planaria with Bifidobacterium longum, a probiotic, will lead to increased clustering behavior, indicative of reduced anxiety in these organisms. Surprisingly, the behavioral assays yielded an opposite outcome. Planaria exposed to the probiotic exhibited heightened anxiety levels and increased locomotion. This unexpected result suggests that the introduction of the foreign probiotic induced dysbiosis in the planaria, disrupting the microbial balance within their system. To further support this theory, dysbiosis was induced in the planaria using a combination of broad-spectrum antibiotics. Ofalaxin, tetracycline, and azithromycin were administered at a concentration of 0.1 mg/ml. Following antibiotic treatment, the behavioral assay was conducted, revealing a phenotype similar to that observed in the probiotic-exposed planaria, including anxiety-like behavior. These findings

provide evidence that Bifidobacterium may induce dysbiosis in planaria, resulting in the manifestation of an anxiety-like phenotype. In conclusion, the administration of Bifidobacterium to planaria led to dysbiosis and the subsequent development of anxiety-like behavior. The use of a combination of broad-spectrum antibiotics, including ofalaxin, tetracycline, and azithromycin, at a concentration of 0.1 mg/ml, successfully induced dysbiosis in the planaria. These observations highlight the intricate interplay between probiotics and host organisms, emphasizing the need for further investigation to elucidate the underlying mechanisms involved. This study contributes to our understanding of the potential impact of Bifidobacterium on planarian behavior and underscores the significance of microbial balance in modulating host phenotypes.

Disclosures: **M. Dodge:** None. **M. Daniels:** None. **A. Reynolds:** None. **M. Scotto:** None. **J. Lichaa:** None. **S. Guariglia:** None.

Poster

PSTR121. Autism: Behavioral and Neural Mechanisms

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR121.03/B17

Topic: A.07. Developmental Disorders

Support: Saint Joseph by the Sea HS

Title: Effects of Lead (Pb2+) Exposure on Planarian Brain Development and Neurobehavioral Phenotypes

Authors: ***M. SCOTTO**[1], J. J. MERLINO[1,2], A. MARANDOLA[1], J. EIDLISZ[3], K. W. HALWANI[3], I. MASSARO[1], A. BIMBO-SZUHAI[1], C. P. CORBO[1,4], S. GUARIGLIA[1,2]; [1]New York State Inst. for Basic Res., Staten Island, NY; [2]St. Joseph by the Sea High Sch., Staten Island, NY; [3]CUNY Col. of Staten Island, Staten Island, NY; [4]Jacksonville Univ., Jacksonville, FL

Abstract: Lead (Pb2+) exposure during early development has been linked to neurodevelopmental disorders. To examine the impact of Pb2+ rapidly on brain development, we conducted a study focusing on neurobehavioral changes, brain anatomy, and molecular pathways in planaria. Planarians were decapitated and exposed to various Pb2+ concentrations (0.1 ppm, 0.25 ppm, and 0.5 ppm) for seven days. After seven days, half of the group remained exposed to Pb2+ until day 14, while the other half was not exposed. At day 7, planarians exposed to 0.5 ppm Pb2+ displayed significant behavioral changes, including increased locomotor activity, enhanced exploration, social clustering, and impaired learning in a passive avoidance task. Through confocal microscopy, we observed reduced gray matter volume in all Pb2+ exposed groups, with the 0.5 ppm group exhibiting the most significant decrease. Cell death, as indicated by cleaved caspase-3 labeling, was notably reduced in all Pb2+ groups compared to the control, suggesting behavioral changes were not primarily caused by cell loss. Moreover, H3 immunostaining

revealed increased cellular proliferation in the central nervous system (CNS) of the 0.5 ppm group, which may contribute to the hyperactive phenotype and abnormal exploration. Animals continuously exposed to Pb2+ for 14 days did not exhibit significant behavioral differences compared to day 7. The 0.5 ppm group maintained the hyperactive phenotype, increased exploration, social clustering, and poor performance in the passive avoidance task. Anatomically, gray matter reduction became more pronounced after 14 days, accompanied by minimal cell death in all Pb2+ exposure groups. The 0.5 ppm group still displayed higher cellular proliferation in the CNS, suggesting its involvement in the hyperactive phenotype. In the groups where Pb2+ exposure was stopped, the 0.5 ppm group retained the hyperactive phenotype and learning impairments. Interestingly, planarians in lower Pb2+ concentration groups, despite lacking behavioral phenotypes, also showed hyperactivity, increased exploration, and cognitive impairments similar to the 0.5 ppm group. These planarians exhibited significantly reduced gray matter volume and increased cellular proliferation, indicating compensatory mechanisms after Pb2+ withdrawal. Gray matter reduction resembling that of persistent Pb2+ treatment groups was anatomically observed but with greater cellular proliferation and no changes in cell death. Our findings suggest that Pb2+ exposure induces a persistent behavioral phenotype in planarians, along with anatomical and cellular changes relevant to understanding Pb2+ toxicity in humans.

Disclosures: **M. Scotto:** None. **J.J. Merlino:** None. **A. Marandola:** None. **J. Eidlisz:** None. **K.W. Halwani:** None. **I. Massaro:** None. **A. Bimbo-Szuhai:** None. **C.P. Corbo:** None. **S. Guariglia:** None.

Poster

PSTR121. Autism: Behavioral and Neural Mechanisms

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR121.04/B18

Topic: A.07. Developmental Disorders

Support: USRG BP 2023
NIGMS- GM113109

Title: Voluntary Exercise Impacts Cognitive Performance in FMR1 KO Rats in an Activity-Dependent Manner

Authors: E. WARNES[1], C. KING[1], A. E. PAHUA[1], M. WILLS[1], C. CROUCH[1], *B. PLAKKE[1], *B. PLAKKE[2];
[2]Psychological Sci., [1]Kansas State Univ., Manhattan, KS

Abstract: Fragile X Syndrome (FXS) is a pervasive neurodevelopmental disorder that is the largest single genetic cause of intellectual disability. FXS is caused by a copy-number repeat expansion mutation in the FMR1 gene, which encodes the FMR protein. The FMR protein is critical in many neurodevelopmental processes, including spatiotemporal regulation of dendrite development. Exercise has shown great promise to bolster neurodevelopmental processes, but

the mechanisms by which it may do so are unclear. The lab's previous work in the valproic acid (VPA) model of autism has shown that exercise can bolster cognitive performance on an attentional set-shifting task. To examine a similar paradigm within the FXS knockout rat, this pilot project placed 11 male Sprague-Dawley rats in a voluntary exercise intervention where they were supplied access to a running wheel for two hours daily for four weeks (n=5 sedentary, n=6 exercise). At the end of the four-week exercise intervention, adult rats underwent a reversal learning task consisting of a compound discrimination phase followed by three reversal phases. There was no effect of condition (sedentary or exercise) on overall task performance (p=0.99). Regression analysis found a relationship between average distance run on the wheel each day and cognitive performance on the compound discrimination phase of the task in the exercise group (p=0.065, r^2=0.61). No effects were found for the reversal phases, suggesting that the benefits of the exercise intervention may be acting on specific and limited neural circuitry. These results suggest that exercise interventions may improve cognitive performance in the FMR1 KO model and supplement the lab's previous findings in the VPA model.

Disclosures: **E. Warnes:** None. **C. King:** None. **A.E. Pahua:** None. **M. Wills:** None. **C. Crouch:** None. **B. Plakke:** None. **B. Plakke:** None.

Poster

PSTR121. Autism: Behavioral and Neural Mechanisms

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR121.05/B19

Topic: A.07. Developmental Disorders

Support: P20GM103418- DRPP to Dr. Bethany Plakke
NIGMS- GM113109 NIH
Start up funds from K-State to Dr. Bethany Plakke

Title: Exercise mitigates brain volume and improves cognitive performance in ASD rodent model

Authors: *A. PAHUA[1], C. KING[1], I. MALI[1], M. PAYNE[1], S. H. BOSSMANN[2], B. PLAKKE[1]; [1]Kansas State Univ., Manhattan, KS; [2]Cancer Biol., Univ. of Kansas Med. Ctr., Kansas City, KS

Abstract: Autism Spectrum Disorder (ASD) has an estimated prevalence of 2.8% in the United States population, with every 1 in 44 children diagnosed as early as 12 months. Core symptoms of ASD are impairments in social communication and restricted repetitive patterns of behaviors, sensitivity to change, and high anxiety levels. Previous research has suggested that exercise mitigates cognitive and social behaviors associated with ASD. The current study analyzed the effect of an aerobic exercise intervention on attentional set-shifting and brain volume in the Valproic Acid rat model of ASD. Pregnant Long-Evans dams received a single dose intraperitoneal injection of either saline or VPA (600 mg/kg valproic acid) on gestational day 12.5. Starting on PND 40, animals were run using a rodent treadmill for 30 minutes/day, 5

days/week for 4 weeks. During the fourth week of exercise, attentional set-shifting was conducted, followed by 3D MRI scans. Blind-to-condition researchers conducted all behavioral data collection and brain segmentation. The data were separated by sex based on past findings. *N*=91 (Male: Exercise Controls 8, Sedentary Controls 10, VPA Exercise 12, VPA Sedentary 13, Female: Exercise Controls 13, Sedentary Controls 10, VPA Exercise 12, VPA Sedentary 13). The exercise intervention improved cognitive performance in the attentional set-shifting task in both VPA and control animals. Results indicate that exercise altered amygdala volumes in males. An effect of sex was also found in the hippocampus and whole brain volume. Previous data has implied that the overgrowth of Lobule VI impairs cognition; however, preliminary data from the current study indicates that exercise modulated the overgrowth and improved cognition. In addition, while analyzing error probabilities (preservative and regressive), it was found that female VPA rats had a higher rate of reoccurring preservative errors than males. These results align with previous findings, indicating that error rates, cognitive performance, and brain volume are mitigated by exercise and may improve cognition in ASD.

Disclosures: **A. Pahua:** None. **C. King:** None. **I. Mali:** None. **M. Payne:** None. **S.H. Bossmann:** None. **B. Plakke:** None.

Poster

PSTR121. Autism: Behavioral and Neural Mechanisms

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR121.06/B20

Topic: A.07. Developmental Disorders

Support: NIH P20 GM103418
NIGMS GM113109

Title: Hippocampal Enlargement Emerges in Adolescence in the Valproic Acid Model and Parallels Human Findings

Authors: *C. KING, H. STRATING, M. PAYNE, I. MALI, S. BOSSMAN, B. PLAKKE; Kansas State Univ., Manhattan, KS

Abstract: Hippocampal enlargement is commonly observed in adolescents with autism spectrum disorder (ASD). fMRI studies indicate that this enlargement may compensate for dysfunctions in the posterior medial network, which is utilized for memory tasks. This study used the valproic acid (VPA) model was used to model ASD in rodents. Our previous work found VPA exposure induced cerebellar pathology in adulthood and caused enlargement of the anterior cingulate cortex (ACC) in female adolescent rats. Some of these enlargements were associated with impaired cognitive performance on an attentional set-shifting task. The current work examined volumetric dysregulation in the cerebellum and hippocampus throughout adolescence. Pregnant Long-Evans dams were given intraperitoneal injections of 600 mg/kg sodium valproate (VPA) or vehicle control at gestational day 12.5. Rats were reared and aged to postnatal day (P)

28 (n=41, 25:16 VPA:Control) or P40 (n=36, 17:19 VPA:Control), which correspond to early and middle adolescence, respectively. At P28 or P40, animals were euthanized by phenobarbital overdose and transcardially perfused with 0.9% saline followed by 4% PFA. Heads were then postfixed in 4% PFA prior to MRI acquisition. After MRI images were obtained, they were segmented in ITK snap for total hippocampus and cerebellum volumes by blind-to-condition researchers. Cerebellar lobules Crus I and Lobule VI were also segmented. Structure volumes were normalized to total brain volume and total cerebellar volume for cerebellar lobules. Data were analyzed by region as normalized volumes in a mixed effects model with litter as a random effect to control for the litter effect when collapsed across sex.
The hippocampus was not enlarged in VPA animals relative to controls at P28 (left p=0.70, right p=0.25), but significant enlargement was observed bilaterally in VPA animals at P40 (left p=0.002, right p=0.03). When separated by sex, both sexes had enlarged left hippocampi (male p=0.06, female p=0.008) and right hippocampi (male p=0.069, female p=0.068). No significant volumetric changes were observed in the cerebellum, but a trend-level increase in left Crus I (normalized to total cerebellum, p=0.079), which is functionally connected to the left hippocampus, was observed. These results show that the VPA model captures specific brain volumetric changes and pathology that parallel human ASD.

Disclosures: C. King: None. **H. Strating:** None. **M. Payne:** None. **I. Mali:** None. **S. Bossman:** None. **B. Plakke:** None.

Poster

PSTR121. Autism: Behavioral and Neural Mechanisms

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR121.07/B21

Topic: A.07. Developmental Disorders

Support: HAWK-IDDRC P50 HD103556
Roy J. Carver Chair in Neuroscience
SFARI grant 345034

Title: Sex-specific neurobiological impact of 16p11.2 hemi-deletion on corticostriatal circuits

Authors: *J. KIM[1], Y. VANROBAEYS[1], B. KELVINGTON[1], C. C. ANGELAKOS[2], J. F. LYNCH, III[1], T. NICKL-JOCKSCHAT[1], T. ABEL[1];
[1]Univ. of Iowa, Iowa City, IA; [2]Psychiatry and Behavioral Sci., Stanford Univ., Stanford, CA

Abstract: Deletion of the 16p11.2 region (16p11.2 del/+) is a copy number variation linked to neurodevelopmental disorders (NDDs). Mice modeling this deletion exhibit alterations in NDD-relevant behaviors linked to corticostriatal circuits. Corticostriatal circuits diverge into the direct and indirect pathways which are characterized by the class of spiny projection neuron (SPN) contacted in the striatum. These pathways are known to have unique molecular properties and unique functions in mediating behavior. We investigated the cell-type specific impact of 16p11.2

deletion on the transcriptome of direct and indirect SPNs. snRNA-seq uncovered striking sex-specific and cell type-specific transcriptomic changes. We then investigated the effect of conditional 16p11.2 del/+ within each SPN subtype on behavior paradigms linked to corticostriatal circuits. This approach yielded distinctive sex-specific behavioral alterations in home-cage activity and reward learning. Together, these results indicate that direct and indirect SPNs differentially contribute to NDD-relevant, sex specific behavioral phenotypes in 16p11.2 del/+ mice. This work lays the foundation for identifying cell type-specific molecular mechanisms mediating corticostriatal circuit dysfunction and male vulnerability or female resilience to NDDs.

Disclosures: **J. Kim:** None. **Y. Vanrobaeys:** None. **B. Kelvington:** None. **C.C. Angelakos:** None. **J.F. Lynch:** None. **T. Nickl-Jockschat:** None. **T. Abel:** None.

Poster

PSTR121. Autism: Behavioral and Neural Mechanisms

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR121.08/B22

Topic: A.07. Developmental Disorders

Support: NIH Grant R01 MH087463
NIH Grant P50 HD103556
NIH Grant T32 GM067795
NIH Grant F31 MH134542

Title: Complement C3 upregulation in the striatum of the 16p11.2 hemi-deletion mouse model contributes to hyperactive behavior

Authors: *B. KELVINGTON, J. KIM, M. E. GAINE, T. ABEL;
Univ. of Iowa, Iowa City, IA

Abstract: The complement system is a major component of the innate immune system and plays an important role in immune surveillance. Recent research has demonstrated that the complement system also plays pivotal roles in brain development, and its dysregulation is involved in neurodegenerative and neuropsychiatric disorders. However, the mechanisms by which the complement system contributes to neurodevelopmental disorders (NDDs) remain poorly understood. In this study, we found that the expression of complement, including complement component 3 (C3), is upregulated in the striatum of mice modeling the 16p11.2 hemi-deletion (16p11.2 del). 16p11.2 hemi-deletion is among the most common copy number variations and results in a high penetrance of NDDs. Additionally, we found that pharmacological inhibition of the C3a receptor alleviates hyperactivity in 16p11.2 del mice, suggesting that elevated complement contributes to NDD-relevant behavior in 16p11.2 del mice. We then profiled the inflammatory state of the 16p11.2 del striatum, a key neural substrate for locomotor behavior, and found that several inflammatory factors are upregulated. Collectively,

these results indicate that increased expression of the complement system, especially C3, mediates hyperactive behavior and is associated with a pro-inflammatory environment in the striatum of 16p11.2 del mice. Our results also suggest that inhibition of an overactive complement system may be an effective strategy to ameliorate NDD symptoms resulting from 16p11.2 hemi-deletion.

Disclosures: B. Kelvington: None. **J. Kim:** None. **M.E. Gaine:** None. **T. Abel:** None.

Poster

PSTR121. Autism: Behavioral and Neural Mechanisms

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR121.09/B23

Topic: A.07. Developmental Disorders

Support: LECOM SEED grant

Title: Abnormal development between the cortical plate and coriticothalamic tract in the embryonic brains of BTBR mice

Authors: *J. LEE, B. BATAZHAN, M. AWAN;
Lake Erie Col. of Osteo. Med., Lakewood Ranch, FL

Abstract: Autism spectrum disorder (ASD) is a neurodevelopmental disorder affecting both the cognitive and behavioral functioning in affected individuals manifested as asocial and repetitive behaviors with limited verbal communication. Histological studies showed that severely affected ASD individuals have enlarged amygdala, hippocampus and cingulate cortex while having lower cell packing density in cortical layers. Development of cortical layers and the corticothalamic tract play very important role in information processing and exchange between various brain regions for establishment and execution of complex behaviors. Our study examined the developmental sequence of cortical plate and corticothalamic tract in the embryonic brains of BTBR mice, an animal model for idiopathic ASD. Embryonic brains of BTBR and control C57 were harvested at E16, E18, E20. Brains were fixed in paraformaldehyde and processed for TBR1 and KI-67 immunofluorescent labeling. Additional brains were transected at mid sagittal plane, DiI crystals were placed in the dorsal thalamic nuclei and fiber labeling was visualized 10 days later. BTBR mice had significantly reduced number of KI-67-positive cells in cortex than C57 at E16 and E18. The reduction was mostly noted in the VZ and SVZ. The TBR1-positive layer was significantly larger in BTBR but the cell packing density was lower than control C57. Such layer differences were not observed at E20. DiI labeling showed that BTBR had significantly more labeled fibers forming the corticothalamic tract than the C57. The DiI-positive fibers from the dorsal thalamus passed thru the caudate/putamen forming the internal capsule and terminated in deeper layers of the cortical plate. Overall E16 and E18 BTBR brains had more numerous DiI-positive fibers in the cortical plate than C57. Our study showed that abnormal formation of embryonic cortical layers and the corticothalamic tract may be associated with the

development of ASD-like phenotype. Understanding the abnormal activation of transcription factors in BTBR may provide insights into the etiology of ASD.

Disclosures: J. Lee: None. **B. Batazhan:** None. **M. Awan:** None.

Poster

PSTR121. Autism: Behavioral and Neural Mechanisms

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR121.10/B24

Topic: A.07. Developmental Disorders

Title: Dha supplementation moderates absolute and relative brain volumes in a prenatal stress-associated asd murine model

Authors: *N. AHMED[1], T. WOO[3], C. KING[1], M. CORDES[1], J. ELLEGOOD[4], J. P. LERCH[5], M. J. WILL[2], D. Q. BEVERSDORF[6];
[2]Dept. of Psychological Sci., [1]Univ. of Missouri, Columbia, Columbia, MO; [3]Dept. of Mol. Pathobiology, New York Univ., New York City, NY; [4]Hosp. For Sick Children, Toronto, ON, Canada; [5]Wellcome Ctr. for Integrative Neuroimaging, Univ. of Oxford, London, United Kingdom; [6]Univ. of Missouri Columbia, Columbia, MO

Abstract: Autism spectrum disorder (ASD) is a neurodevelopmental condition characterized by social communication difficulties, restricted interests, and repetitive behaviors. Gene-environment interactions appear to contribute to the pathogenesis. One exemplar involves prenatal stress exposed pregnancies in mothers with at least one copy of the short allele in the promotor region of the serotonin transporter gene (SERT), modeled using dams with the heterozygous serotonin transporter knockout (SERT +/-) in mice, whose offspring produce ASD-associated phenotypical behaviors of low social ability and increased repetitive grooming. Our previous work showed that in the maternal SERT +/- prenatal stress-associated ASD mouse model, DHA supplementation mitigates dopaminergic excess found in the striatum of this model, as well as the behavioral effects. The present study aimed to quantify the absolute and relative brain volume variability across offspring of SERT +/- dams exposed to prenatal stress vs C57BL/6J controls with and without DHA supplementation initiated before pregnancy in the dams and continuing through weaning. After weaning, offspring of those given supplementation also received DHA until behavioral testing began. Brains were collected from offspring around post-natal day 70. Using deformation-based morphometry at 7T MRI, we assessed neuroanatomical differences across our four conditions (SERT +/- prenatally stressed with DHA supplementation(HS/D), SERT +/- prenatally stressed with no supplementation (HS/ND), C57BL/6J not stressed with DHA supplementation (WT/D) , and C57BL/6J not stressed with no supplementation(WT/ND). Without DHA supplementation, SERT +/- prenatally stressed offspring were seen to have significantly larger absolute and relative brain volumes in the striatum, nucleus accumbens, olfactory peduncle, flocculus, claustrum and cingulate cortex areas 24a and 24b compared to wildtype controls. DHA appeared to reverse many of these findings, as

several areas related to the striatum and dopaminergic pathways were significantly decreased in the HS/D compared toHS/ND offspring including the amygdala, thalamus, olfactory peduncle, flocculus, claustrum, and the cingulate cortex areas 24a, 24b, 24b.1, 29a, 29b, 29c, 30, and 32. In contrast, only the olfactory peduncle and the cingulate cortex areas 24a and 24b showed a relative area decrease between SERT +/- stressed offspring controls and those given DHA. In conclusion, DHA supplementation in the prenatal stress-associated ASD mouse model seems to influence a wide variety of brain regions including those associated with the striatum and its related dopaminergic pathways.

Disclosures: **N. Ahmed:** None. **T. Woo:** None. **C. King:** None. **M. Cordes:** None. **J. Ellegood:** None. **J.P. Lerch:** None. **M.J. Will:** None. **D.Q. Beversdorf:** None.

Poster

PSTR121. Autism: Behavioral and Neural Mechanisms

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR121.11/B25

Topic: A.07. Developmental Disorders

Support: JSPS KAKENHI
MEXT KAKENHI
the Asahi Glass Foundation
Naito Foundation
Takeda Science Foundation

Title: Oxytocin restores abnormal social behavior in a mouse model of 3q29 microdeletion.

Authors: *T. TAKEMOTO[1], K. KITAGAWA[1], K. NAGAYASU[2,1], K. SEIRIKI[1], A. HAYATA-TAKANO[1], A. KASAI[1], Y. AGO[3], K. TAKUMA[1], D. MORI[4], N. OZAKI[4], R. HASHIMOTO[5], H. HASHIMOTO[1,5,6], T. NAKAZAWA[1,5,7];
[1]Osaka Univ., Osaka, Japan; [2]Kyoto Univ., Kyoto, Japan; [3]Hiroshima Univ., Hiroshima, Japan; [4]Nagoya Univ., Aichi, Japan; [5]Natl. Ctr. of Neurol. and Psychiatry, Tokyo, Japan; [6]United Grad. Sch. of Child Development, Osaka University, Kanazawa University, Hamamatsu Univ. Sch. of Medicine, Chiba Univ. and Univ. of Fukui, Osaka, Japan; [7]Tokyo Univ. Agri., Tokyo, Japan

Abstract: Autism spectrum disorder (ASD) is a neurodevelopmental condition characterized by specific social impairments, restricted interests, and repetitive and stereotyped behaviors. Given that the 3q29 microdeletion, a recurrent copy number variant, significantly increases a risk for both ASD and schizophrenia, this deletion serves as a valuable pathological model for exploring the molecular and cellular mechanisms of psychiatric conditions. We previously generated a genetically engineered mouse carrying a deletion of the chromosomal region corresponding to the human 3q29 region (Df/+ mice). These mice exhibit behavioral abnormalities associated with psychiatric disorders, suggesting the relevance of Df/+ mice as an effective model for psychiatric disorders with high construct and face validity. In this study, to elucidate the molecular and

cellular mechanisms behind the abnormal social behavior in Df/+ mice, we investigated the possible involvement of oxytocin (OXT) signaling in Df/+ mice, because the OXT system exerts significant influence on social behavior across species. In Df/+ mice, we found that the number of OXT-positive cells was significantly decreased in the paraventricular nucleus, where OXT neurons are densely located. Consistent with this, we demonstrated that the levels of OXT were significantly lower in the cerebral cortex in Df/+ mice. Importantly, we demonstrated that intraperitoneal administration of OXT restored abnormal social behavior in Df/+ mice. Our previous observation showing that relatively high cell excitability in the auditory cortex of Df/+ mice after social interaction, compared to wild-type mice, implies that administration of OXT may suppress the higher cell excitability in the auditory cortex of Df/+ mice. Considering that the precise molecular and cellular mechanisms underlying the behavioral phenotypes in Df/+ mice remain poorly understood, our findings provide valuable insights into the molecular and cellular pathogenesis underlying the 3q29 microdeletion.

Disclosures: T. Takemoto: None. **K. Kitagawa:** None. **K. Nagayasu:** None. **K. Seiriki:** None. **A. Hayata-Takano:** None. **A. Kasai:** None. **Y. Ago:** None. **K. Takuma:** None. **D. Mori:** None. **N. Ozaki:** None. **R. Hashimoto:** None. **H. Hashimoto:** None. **T. Nakazawa:** None.

Poster

PSTR121. Autism: Behavioral and Neural Mechanisms

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR121.12/B26

Topic: A.07. Developmental Disorders

Support: NIH-R01 GM135087

Title: Sensory neocortical circuit dysfunction in a mouse model of Setd2 haploinsufficiency.

Authors: *A. POPE[1], J. RICHTER[2], C. WILLIAMS[3], J. GIBSON[4], K. HUBER[4];
[1]UT Southwestern Neurosci. Grad. Program, Dallas, TX; [2]Univ. Mass. Med. Sch., Amherst, MA; [3]Neurosci., UT Southwestern Med. Ctr., Dallas, TX; [4]Neurosci., Univ. of Texas Southwestern Med. Ctr. at Dallas, Dallas, TX

Abstract: Post-translational modifications of histones, including methylation, are critical for regulating gene expression and maintaining homeostasis. In neurons, dysregulation of histone methylation is associated with neurodevelopmental disorders such as autism spectrum disorders (ASD). SET Domain Containing 2, histone lysine methyltransferase (SETD2) that methylates histone H3 at lysine 36 (H3K36) and is the primary methyltransferase for trimethylating H3K36 in human (H3K36me3). SETD2 is unique among histone methyltransferase in that it directly binds RNA Pol ll and deposits H3K36me3 co-transcriptionally. H3K36me3 primarily involved in transcriptional regulation but can also regulate alternative splicing. Haploinsufficiency of *SETD2* is responsible for Sotos-like overgrowth syndrome, characterized by obesity, macrocephaly, intellectual disability, autism, and epilepsy. *SETD2* is also considered a “high

confidence" ASD-risk gene in the SFARI gene database. Little is known of the role of SETD2 in the brain. There is also little known of the role of histone methylation, specifically H3K36me3, in postnatal developing brain. To address these gaps in knowledge, I made a mouse with conditional deletion of *Setd2* postnatally (~P8) in excitatory forebrain neurons using the BAC-CaMKIIα Cre line. Acute slices of somatosensory neocortex from young, 3rd postnatal week, *Setd2* heterozygous mice (*SetD2* cHet) have increased circuit excitability, as measured by the duration and power of persistent activity, or UP, states. Additionally, in *Setd2* cHet, long duration UP states are rescued by a negative allosteric modulator of metabotropic glutamate receptor 5 (mGluR5), as well as protein synthesis inhibitors. This suggests hyperactivation of the mGluR5 signaling pathway to protein synthesis with *Setd2* deletion. **I hypothesize that loss of SETD2 leads to dysregulation of genes that may encode components of protein synthesis machinery, proteins that regulate synaptic, neuronal and circuit function and/or mGluR5, its interacting partners or effectors**

Disclosures: A. Pope: None. **J. Richter:** None. **C. Williams:** None. **J. Gibson:** None. **K. Huber:** None.

Poster

PSTR121. Autism: Behavioral and Neural Mechanisms

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR121.13/Web Only

Topic: A.07. Developmental Disorders

Support: South Dakota Biomedical Research Infrastructure Network (P20GM103443)
South Dakota EPSCoR (IIA-1355423)
Augustana University Biology Department
National Institute of Mental Health, MH106640 (SSN)

Title: Carbamoylated erythropoietin rescues social defects in a mouse model of autism

Authors: *A. KLOTH[1], S. LEMKE[1], A. STREET[1], L. SCHOENBECK[1], K. SCHUMACHER[1], V. THAKKAR[1], R. BUNDY[1], E. VIEHWEG[1], M. SATHYANESAN[2,3], S. NEWTON[2,3]; [1]Augustana Univ., Sioux Falls, SD; [2]Div. of Basic Biomed. Sciences, Sanford Sch. of Med., Univ. of South Dakota, Vermillion, SD; [3]Sioux Falls VA Hlth. Care Syst., Sioux Falls, SD

Abstract: Autism spectrum disorder (ASD) is a neurodevelopmental disorder that affects over 2% of the population worldwide, which is characterized by repetitive behaviors, restricted areas of interest, deficits in social communication, and high anxiety. Currently, there are no known effective treatments for the core features of ASD. Previous literature has established erythropoietin (EPO) as a promising antidepressant, working as a potent neurogenic and neurotrophic agent with hematopoietic side effects. Carbamoylated erythropoietin (CEPO), a chemically engineered non-erythropietic derivative of EPO, appears to retain the neuroprotective

factors of EPO without the hematologic properties, and recent evidence shows that CEPO corrects stress-related depressive behaviors in BALB/c mice. We investigated whether CEPO can recover deficient social behavior and anxiety-related behavior that give the BALB/c mouse model face validity for ASD when compared to C57BL/6J controls. After administering CEPO (40 μg/kg in PBS) or vehicle over 21 days, we analyzed performance in the three-chamber social approach, the open field, the elevated plus maze, and the Porsolt's forced swim tests. CEPO appears to correct sociability in the three-chamber social approach task to C57 levels, increasing the number of social interactions overall rather than altering the overall amount of exploratory activity in the maze. Consistent with this finding, there was no concomitant increase in distance traveled in the open field, nor was there an alteration of anxiety-related measures in the task. On the other hand, CEPO administration improved exploration in the elevated plus maze. Ongoing experiments will determine whether similar effects occur in female mice under the same dosage regimen and will investigate whether CEPO engages neurotrophic mechanisms and alters neuronal activity in the hippocampus and other areas involved in social behavior.

Disclosures: **A. Kloth:** None. **S. Lemke:** None. **A. Street:** None. **L. Schoenbeck:** None. **K. Schumacher:** None. **V. Thakkar:** None. **R. Bundy:** None. **E. Viehweg:** None. **M. Sathyanesan:** None. **S. Newton:** None.

Poster

PSTR121. Autism: Behavioral and Neural Mechanisms

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR121.14/B27

Topic: A.07. Developmental Disorders

Support: American Psychological Foundation / Council of Graduate Departments of Psychology (APF/COGDOP) Ruth G. and Joseph D. Matarazzo Scholarship
Center for OCD, Anxiety and Related Disorders (COARD) University of Florida Pilot Grant
Jacquelin Goldman Research Grant in Developmental Psychology
Trish Calvert Ring Dissertation Fellowship

Title: Functional connectivity alterations associated with restricted repetitive behavior and its attenuation by environmental enrichment

Authors: *A. L. FARMER[1], M. FEBO[2], B. J. WILKES[3], M. H. LEWIS[2];
[1]Psychology, [2]Psychiatry, [3]Applied Physiol. & Kinesiology, Univ. of Florida, Gainesville, FL

Abstract: Restricted repetitive behaviors (RRB) are inflexible motor and cognitive responses diagnostic for autism and common in neurodevelopmental disorders. As yet, no effective biomedical treatments exist for RRB. Studies using animal models, however, show environmental enrichment (EE) reduces RRB. Identifying underlying mechanisms of EE effects

could yield potential intervention targets. We used functional magnetic resonance imaging (fMRI) to identify functional connectivity differences associated with RRB and its attenuation by EE in male and female C58 mice, an animal model of RRB, and C57BL/6 controls lacking RRB. Mice were randomly assigned to standard or EE housing at weaning. At 6 weeks postweaning, we assessed repetitive motor behavior and acquired anatomical and resting state fMRI scans. These same methods were performed in a younger cohort of male and female C58 mice at 3 weeks postweaning to examine EE effects during RRB development. fMRI data were preprocessed using software tools in Analysis of Functional NeuroImages, FMRIB Software Library and Advanced Normalization Tools, which entailed skull-stripping, susceptibility correction, despiking, motion and linear drift removal, highpass filter, independent component analysis (ICA) noise reduction, correction for field inhomogeneities, and registration to the Allen mouse brain atlas. Separate ICAs were conducted for each age cohort in FSL MELODIC to identify 20 resting state networks (RSN). Analyses were conducted in FSL dual regression using 5000 permutations and threshold-free cluster enhancement. Mouse strain and housing differences were examined with t-tests, and RRB scores were correlated with connectivity in each RSN. Mouse strain differences were found in 10 of the 20 RSNs. In the older cohort, EE increased connectivity between the left somatosensory network and the right hippocampus and pretectal nuclei. In the younger cohort, EE reduced connectivity between the left visual network and right area prostriata and postsubiculum. In the older cohort, RRB was positively correlated with connectivity between the right somatosensory network and right thalamic nuclei and dorsomedial striatum. It was also positively correlated with connectivity in a ventrocaudal striatum network with the right medial and posterior amygdalar nuclei. RRB in the younger cohort was associated with reduced connectivity between the right visual network and right ventral striatum. Our results suggest widespread strain differences, as well as RRB associations with aberrant sensory and pain processing and threat perception. EE may reduce RRB by altering connectivity in the somatosensory and visual networks.

Disclosures: **A.L. Farmer:** None. **M. Febo:** None. **B.J. Wilkes:** None. **M.H. Lewis:** None.

Poster

PSTR121. Autism: Behavioral and Neural Mechanisms

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR121.15/B28

Topic: A.07. Developmental Disorders

Support: CIHR
OBI (Province of Ontario Neurodevelopmental Network)

Title: Longitudinal effects on offspring brain development and behaviour following maternal autoantibody exposure

Authors: *T. CHIEN[1,3], J. MCLELLAN[4], M. R. BRUCE[4], E. ANAGNOSTOU[5], L. SERGHIDES[2,6], J. VAN DE WATER[4], J. G. SLED[1,3], J. P. LERCH[1,3];

[1]Med. Biophysics, [2]Immunol., Univ. of Toronto, Toronto, ON, Canada; [3]Hosp. for Sick Children, Toronto, ON, Canada; [4]MIND Inst., Univ. of California - Davis, Sacramento, CA; [5]Holland Bloorview Kids Rehabil. Hosp., Toronto, ON, Canada; [6]Toronto Gen. Hosp. Res. Inst., Toronto, ON, Canada

Abstract: IgG autoantibodies (aAbs) reactive to fetal brain proteins have been discovered in ~20% of mothers of children with autism spectrum disorders (ASD), yet little is known about their effects on the developing brain and their role in mediating ASD (Jones et al., 2020). Neurodevelopment takes place in distinct stages during the prenatal and early postnatal period in humans and rodents. Therefore, exposure to pathogenic aAbs during the fetal period may have characteristic brain or behavioral outcomes.In this study, we investigated the effects of maternal aAbs on offspring neuroanatomy and behavior through a comprehensive longitudinal analysis. The objective is to determine, potentially sex-linked, neuroanatomical differences that manifest at specific timepoints in response to aAb exposure, and therefore may mediate the stereotypical behaviors in ASD. Maternal aAb related (MAR)-ASD mouse model was generated according to the endogenous antigen-driven model developed by the Van de Water Lab using LDH-A/-B, CRMP1, and STIP1 antigenic peptides (Jones et al., 2020). Male and female C57Bl6/J offspring were used to conduct *in vivo* whole brain manganese enhanced magnetic resonance imaging at 8 timepoints, and a series of behavioral assays to examine characteristic ASD-related behaviors including sociability, communication, hyperactivity, and anxiety. Preliminary neuroanatomical results found a subtle pattern of reduced volume in white matter regions such as the corpus callosum and basal nuclei structures including the basal forebrain at postnatal day 17 in MAR-ASD offspring, but these differences were normalized at adulthood (p65). This pattern was observed across both sexes albeit seems to be more prominent in females. No treatment effect was found on any of the behavior assays. Our results are consistent with previous findings in which individuals with autism have volume loss in the basal ganglia associated with impaired cholinergic innervation to the cortex that may account for cognitive dysfunction and altered social behaviors (Riva et al., 2011). Furthermore, a decrease in choline levels was observed in a recent rat model of MAR-ASD offspring, which correlated with a reduction in cingulate cortex volume compared to controls (Bruce et al., 2023). These results suggest MAR-ASD aAb exposure may affect the cholinergic system of the basal forebrain or myelination in white matter regions. Since choline is a precursor to the neurotransmitter acetylcholine, myelin, and cell membrane lipids, it is plausible that aAbs may affect offspring brain protein function and myelin turnover which could potentially contribute to the altered brain development observed.

Disclosures: T. Chien: None. **J. McLellan:** None. **M.R. Bruce:** None. **E. Anagnostou:** None. **L. Serghides:** None. **J. Van de Water:** E. Ownership Interest (stock, stock options, royalty, receipt of intellectual property rights/patent holder, excluding diversified mutual funds); patent application involving the MAR-ASD peptides described herein and has a UC Davis-based startup company focusing on the development of the MAR-ASD autoantibody profile as a risk assessment. **J.G. Sled:** None. **J.P. Lerch:** None.

Poster

PSTR121. Autism: Behavioral and Neural Mechanisms

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR121.16/B29

Topic: A.07. Developmental Disorders

Support: JSPS KAKENHI Grant 21H05326
JSPS KAKENHI Grant 22H00986
JSPS KAKENHI Grant 23H03883

Title: Crosstalk between gut microbiota and host response in autism associated dyrk1a mutant of zebrafish

Authors: *T. NISHIMURA[1], R. KIMURA[1], S. R. SUZUKI[1], Y. LI[1], S. MAEGAWA[2], M. HAGIWARA[1];
[1]Dept. of Anat. and Developmental Biol., Grad. Sch. of Medicine, Kyoto Univ., Kyoto, Japan;
[2]Dept. of Intelligence Sci. and Technol., Grad. Sch. of Informatics, Kyoto Univ., Kyoto, Japan

Abstract: Gut-brain interaction in autism spectrum disorder (ASD) has been rising attention recently. In fact, many ASD patients suffer from gastrointestinal (GI) problems including constipation and diarrhea. However, few animal model studies have examined interactions between gut microbiota and host response in ASD. In this study, we focused on *DYRK1A*, one of the high-confidence ASD risk genes, known to be associated with GI symptoms. CRISPR/Cas9 was applied to generate a *dyrk1aa* loss-of-function mutation (*dyrk1aa*$^{-/-}$) in zebrafish. To reveal interactions between gut microbiome and host response in *dyrk1aa* knock-out (KO) zebrafish, morphological measurements, transcriptome, and microbiome analyses were performed. The analysis of the GI tract of *dyrk1aa* KO zebrafish revealed that significantly altered gut morphology. In particular, *dyrk1aa* KO zebrafish showed a significantly decreased lower villi and the fewer goblet cells. To get further insight into molecular mechanisms underlying these findings, we performed RNA sequencing on intestine samples. This approach identified 402 significantly up-regulated genes in *dyrk1aa* KO zebrafish (FDR < 0.05, fold change > 2). Interestingly, these genes were significantly associated with proteolysis and immune system. To reveal the microbial diversity and abundance, the Illumina MiSeq sequencing of the V3-V4 region of the 16S rRNA gene was conducted. As expected, we found that low richness in gut microbiota from *dyrk1aa* KO zebrafish. As for the microbial composition, Acidimicrobiia, Bacteroidia, and Babeliae were decreased, Gammaproteobacteria was increased in *dyrk1aa* KO zebrafish compared with wild-type. Overall, these results provide the evidence of interactions between dysfunction of gut and dysbiosis in *dyrk1aa* KO zebrafish. Our findings could help to further understand the gut-brain interaction in ASD.

Disclosures: T. Nishimura: None. **R. Kimura:** None. **S.R. Suzuki:** None. **Y. Li:** None. **S. Maegawa:** None. **M. Hagiwara:** None.

Poster

PSTR122. Animal Models of Behavioral Dysfunction

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR122.01/B30

Topic: A.09. Adolescent Development

Support: NIH Grant RO1

Title: Neuronal heterogeneity in the mouse paralaminar nucleus

Authors: *V. A. BUTYRKIN[1,2], D. SAXON[3,2], P. J. ALDERMAN[5], S. F. SORRELLS[5], S. VICINI[4], J. CORBIN[2];
[1]Neurosci. and Cognitive Sci. Program (NACS), Univ. of Maryland, Col. Park, College Park, MD; [2]Ctr. for Neurosci. Res., Children's Natl. Hosp., Washington, DC; [3]Georgetown Univ. Med. Center, Interdisciplinary Program In Neurosci., [4]Dept. of Pharmacol. and Physiol., Georgetown Univ. Med. Ctr., Washington, DC; [5]Dept. of Neurosci., Univ. of Pittsburgh, Pittsburgh, PA

Abstract: The amygdala is the central structure of the limbic system and is responsible for imparting salience to environmental cues. While the neurons of the amygdala are generated from embryogenesis, the amygdala undergoes a protracted growth period through adolescence. Previous studies from our group and others revealed that this growth, in large part, is contributed by changes in the amygdala paralaminar (PL) nucleus. Our work has shown that a hallmark feature of the PL is that it contains a group of prenatally born immature neurons that mature throughout the first two decades of life in humans. Thus, impairment of PL neuronal maturation can have consequences for neuropsychiatric disorders that manifest in childhood and adolescence, including anxiety, bipolar disorder, schizophrenia, and autism. Our recent research has revealed the presence of a similar population of neurons in the mouse amygdala. Our electrophysiological and morphological studies revealed that PL neurons comprise two different types of excitatory (Tbr1+) neurons. Based on these data, we hypothesized that late-maturing neurons in the PL maybe generated from different embryonic progenitor populations. To test this, we fate-mapped PL neurons using Emx1CreRYFP and Gsx2iCreRYFP, mice which broadly mark pallial (cortical) and subpallial (subcortical) progenitor populations in the embryonic brain. We found that each lineage contributes to the diversity of PL neurons in interesting patterns. We are currently further exploring the developmental trajectory of these neurons from their birth in the embryonic brain, their migratory routes, and integration into PL functional circuitry.

Disclosures: V.A. Butyrkin: None. **D. Saxon:** None. **P.J. Alderman:** None. **S.F. Sorrells:** None. **S. Vicini:** None. **J. Corbin:** None.

Poster

PSTR122. Animal Models of Behavioral Dysfunction

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR122.02/B31

Topic: A.09. Adolescent Development

Support: NIH Grant R01-MH086507
NIH Grant R01-MH127850

Title: Disruption of pre-pulse inhibition of the acoustic startle following early life adversity

Authors: *E. ARTUR DE LA VILLARMOIS[1], H. BRENHOUSE[2], K. Y. TSENG[1];
[1]Univ. of Illinois Chicago, CHICAGO, IL; [2]Northeastern Univ., Boston, MA

Abstract: Early life adversity (ELA) is associated with developmental dysregulations of corticolimbic circuits including the prefrontal cortex (PFC) and the basolateral amygdala (BLA). Such neural circuit disruption is thought to trigger the onset of maladaptive cognitive and anxiety-like behaviors. Here we used an age-sensitive pre-pulse inhibition (PPI) of the acoustic startle response to reveal if exposure to maternal separation ELA can lead to changes in the inhibition of the pre-pulse. The PPI behavioral construct was chosen because is not entirely PFC- or BLA-dependent, but sensitive to local changes in GABA and glutamate function, and often used to reveal dysregulation of cognitive and anxiety-like responses. We found that the level of PPI increases through adolescence to reach adulthood level by P70, however rats exposed to maternal separation ELA from postnatal days (P) 2 to 20 exhibit a PPI deficit in adulthood. Interestingly, a similar PPI deficit was observed in male and female rats exposed to a shorter ELA period (from P11 to P20). It is conceivable that ongoing development processes within P11-P20 window are critical for the maturation of the PPI response. Ongoing experiments will reveal the impact of maternal separation ELA from P2 to P10 to determine whether there is a critical developmental window of susceptibility to ELA.

Disclosures: E. Artur De La Villarmois: None. **H. Brenhouse:** None. **K.Y. Tseng:** None.

Poster

PSTR122. Animal Models of Behavioral Dysfunction

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR122.03/B32

Topic: A.09. Adolescent Development

Support: BSF-NSF-2021-776

Title: Behavioral and neuronal signatures of adolescence in the mouse auditory cortex

Authors: *B. PRAEGEL[1], A. DYM[1], F. CHENG[2], S. DRUCKMANN[2], A. MIZRAHI[1];
[1]Hebrew Univ. of Jerusalem, Jerusalem, Israel; [2]Stanfort Univ., Stanford, CA

Abstract: Adolescence is known to be a period of uncertainty, exploration, and learning. Our understanding of the underlying neural correlates of adolescence remains scarce. Here, we studied adolescence through the prism of auditory learning, and the neural representations of learned sounds in the auditory cortex of mice. We asked whether adolescent and adult mice discriminate tone categories differently, and how are these differences expressed in auditory

cortical responses in behaving mice. First, we trained freely behaving mice (n=15 adult mice and n=15 adolescent mice) to perform a go no-go task of pure tone categories. We reveal weaker performance in adolescence compared to adulthood and found that it was attributed to specific biases. Second, we trained head-restrained mice (n=6 adult mice and n=5 adolescent mice) on the same task and performed two separate experiments: 1) We manipulated auditory cortex on a trial-by-trial basis using optogenetic silencing (n = 4 mice injected with the GtACR2 opsin and n = 2 control mice inject with AAV-CAMKII-GFP). Inhibiting auditory cortex in adult mice decreased performance, indicating a causal relationship between auditory cortex and tone categorization. 2) We recorded single units in the auditory cortex during engaged behavior using neuropixels (n=14 adult recordings and n = 13 adolescent recordings). We isolated units from primary auditory cortex, secondary auditory cortex, and temporal association area (n = 1174 units in adult mice and n = 1134 units in adolescent mice). We are currently evaluating the task-, stimulus- and choice-related activity in single neurons, as well as in population dynamics. These data will allow us to reveal the neural correlates of behavior in adolescence as compared to adulthood.

Disclosures: B. Praegel: None. **A. Dym:** None. **F. Cheng:** None. **S. Druckmann:** None. **A. Mizrahi:** None.

Poster

PSTR122. Animal Models of Behavioral Dysfunction

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR122.04/B33

Topic: A.09. Adolescent Development

Title: Prior experience does not markedly affect evoked activity in the amygdala of young rats exploring a space with or without objects

Authors: R. M. SOUDERS[1], T. M. STRONG[1], R. J. MOE[1], ***M. C. ZRULL**[2];
[1]Psychology, [2]Appalachian State Univ., Boone, NC

Abstract: Adolescence is often marked by stimulation seeking and emotion-driven behavior, both of which can lead to risk-taking behavior. Exposure to a new or known environment can promote exploration and emotional response through sensory and motor stimulation, provide opportunity to interact with same-sex conspecifics, and allow investigation of objects and the space itself. Given the role of the amygdala in emotional response and evaluation of environmental and social cues, we examined how exposure to a unique space with or without various moveable objects might evoke neural activity across lateral (LA) and basolateral (BLA) amygdala in rats with or without a history of experience in that particular space. We used c-FOS to study the effects of the experience on evoked neural response in the LA and BLA of 46 adolescent rats. After weaning, the rats were housed in groups in standard shoebox cages and spent 1.5 h, 5 days/week in an enclosure with ramps and platforms with (EEob) or without (EEno) objects or were only handled (No group). At 49 days old and prior to sacrifice, rats from

each experience group spent 1.5 h in the enclosure with (NoEEob, EEobEEob, EEnoEEob groups) or without (NoEEno, EEobEEno, EEnoEEno) or in a standard cage in a quiet and dark room (controls, NoNo, EEobNo, EEnoNo). Brains were processed to visualize c-FOS+ neurons, and cell counts were made using digital microscopy and stereological technique. Data were standardized to NoNo group cell counts. For LA and BLA, any exposure to a space, whether with or without objects and without regard to prior experience in a similar setting, increased neural activity over no exposure (NoNo controls), +52% ($p=.004$) and +44% ($p<.001$), respectively. For LA, the effect was driven by NoEEob and NoEEno increased activity over NoNo rats (+57%, $p=.007$) without significant contribution from prior history conditions. For BLA, both rats without prior history (NoEEob, NoEEno, +35%, $p=.014$) and with prior exposure to the space without objects (EEnoEEob, EEnoEEno, +76%, $p<.001$) showed more activated neurons than specific controls (NoNo, EEnoNo). Our results imply, in general, exposure to a familiar or novel environment with or without objects promotes activity in LA and BLA that is a likely contributor to behavior and exploration in response to the current situation. For the adolescent rats in this study, prior experience with other rats in a stimulating environment, which contained objects or not, did not appear to have substantial impact on neural responses in the amygdala when faced with the opportunity to interact with other rats and investigate that unique environment again.

Disclosures: R.M. Souders: None. **T.M. Strong:** None. **R.J. Moe:** None. **M.C. Zrull:** None.

Poster

PSTR122. Animal Models of Behavioral Dysfunction

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR122.05/B34

Topic: A.09. Adolescent Development

Support: Internal Funds

Title: The Influence of Puberty on Risk-Taking and Reward-Seeking Behaviors.

Authors: *H. RUBY, V. RIESGO, M. CZUBA, A. GONZALEZ, J. WILLING;
Bowling Green State Univ., Bowling Green, OH

Abstract: The Influence of Puberty on Risk-Taking and Reward-Seeking Behaviors.**AuthorsHallie Ruby***, Victoria Riesgo, Madeline Czuba, Antonio Gonzalez, Jari Willing**Abstract**Adolescence, defined loosely as the transitionary period between childhood and adulthood, is a crucial time for neurodevelopment and is marked by the achievement of sexual maturation, also known as puberty. One of the brain regions affected by puberty is the prefrontal cortex, in which pubertal onset coincides with a decrease in the number of neurons and synapses. Furthermore, puberty influences behaviors by the prefrontal cortex including studies that found postpubertal individuals of both sexes demonstrated an increase in cognitive flexibility. Similar to the previously mentioned study, the present study aimed to investigate the connection between

pubertal onset and PFC-regulated behaviors, particularly the regulation between risk and reward. Using physical determinants of pubertal onset, our behavioral study divided subjects into prepubertal and postpubertal groups and engaged them in a risk-taking test that required individuals to pass from a start chamber through a chamber with bedding soaked in predator odor to acquire a sucrose pellet reward in the third chamber. We found that post-pubertal males spent less time in the start chamber during training and consumed the reward earlier than prepubertal males during training. Additionally, in our preliminary analysis, we discovered a significant sex-by-pubertal interaction between the number of pellets consumed and time spent in the second chamber. The results of these studies support the notion that adolescents are more likely to participate in risky behavior after puberty. Furthermore, our findings suggest that pubertal effects on risky behavior during adolescence are sex-specific.

Disclosures: **H. Ruby:** None. **V. Riesgo:** None. **M. Czuba:** None. **A. Gonzalez:** None. **J. Willing:** None.

Poster

PSTR122. Animal Models of Behavioral Dysfunction

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR122.06/B35

Topic: A.09. Adolescent Development

Support: National Institute of Child Health and Human Development HD109618-01A1

Title: Adolescent Hormonal Contraceptive Administration Impacts Prefrontal Cortex Myelin Basic Protein and Risk-Assessment Behavior in Female Rats.

Authors: *R. A. GILFARB[1], S. S. RANADE[3], M. STEWART[3], A. RAJESH[3], C. DYE[3], K. M. LENZ[4], B. LEUNER[2];
[1]Psychology, [2]Psychology Dept., Ohio State Univ., Columbus, OH; [3]Psychology Dept., [4]Psychology, The Ohio State Univ., Columbus, OH

Abstract: Hormonal contraceptives (HCs) are commonly used during adolescence, despite their unknown effects on brain and behavioral maturation. The prefrontal cortex (PFC) is a region that continues to develop structurally and functionally throughout adolescence. Some aspects of PFC development, such as myelination, are mediated by the same hormones that HCs target. As such, we hypothesize that HC exposure during the adolescent window of brain development may disrupt PFC myelination to influence behavior. To test this hypothesis, intact female Sprague-Dawley rats were randomly assigned to receive daily subcutaneous injections of either vehicle or ethinyl estradiol + levonorgestrel (HC) for the duration of adolescence (postnatal day 35-56). Estrous cycles were monitored throughout the treatment. Blood for ELISA and brain tissue for liquid chromatography/mass spectrometry (LC-MS) or immunohistochemistry were collected and behavioral assays performed between postnatal day 57 and 63, with all analyses completed

blind to experimental condition. HC treatment was effective, with vaginal lavage indicating disrupted estrous cycling and ELISA quantification showing diminished luteinizing hormone concentration in HC-treated rats (n = 9-10/group). LC-MS revealed that EE and LNG were present in the brain following HC treatment (n = 3-4/group). Percentage area immunolabeling for myelin basic protein (MBP) in the medial prefrontal cortex (mPFC) was increased in HC-treated rats while no differences in density of MBP staining were detected (n = 11-12/group). Microglia can influence myelination, thus the percentage area of mPFC Iba1 (microglia) immunolabeling was also assessed and found to be reduced in HC-treated rats (n = 10-12/group). Finally, behavioral assessments showed that HC treatment diminished risk-assessment behavior in the elevated plus maze (n = 19/group) and novelty-induced hypophagia paradigm (n = 11-13), which may suggest that HC administration during adolescence prevents the typical development of impulse inhibition. Overall, these studies provide some of the first evidence to demonstrate that one of the most widely used pharmaceuticals (HCs) given during a vulnerable developmental period (adolescence) influence PFC development, which may contribute to altered risk-assessment behavior.

Disclosures: R.A. Gilfarb: None. **S.S. Ranade:** None. **M. Stewart:** None. **A. Rajesh:** None. **C. Dye:** None. **K.M. Lenz:** None. **B. Leuner:** None.

Poster

PSTR122. Animal Models of Behavioral Dysfunction

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR122.07/B36

Topic: A.09. Adolescent Development

Title: Multidimensional analysis of influence of rearing environment on development in common marmoset.

Authors: *F. SEKI, T. INOUE, T. YURIMOTO, M. TOGASHI, T. MURAOKA, K. MUKASA, J. HOSHI, Y. KOMAKI, Y. KUROTAKI, E. SASAKI;
Central Inst. For Exptl. Animals, Kawasaki-shi, Kanagawa, Japan

Abstract: The rearing environment may affect the physical and brain development of captive marmosets. In this study, we compared marmoset physical and brain growths that were kept in the standard cage (W820×D610×H1600mm, standard group) and enriched environment of a wide room containing natural wooden logs (W 5240×D 3560×H2500 mm, enriched group) with family members.Physical examinations were conducted to investigate growth curves of their body weight and body length. The whole-body was also scanned using CT at the adult age of 1.5-3.0 years. As the results of the physical examinations, there were no significant difference in physical measurements such as growth curves of body weight and length, fat volume, and bone mineral density between standard and enriched group marmosets.For the examination of brain development, longitudinal MRI was conducted from the age of 3-18 months. T1-weighted images were acquired to investigate whether the developmental volume change was different

between enriched and standard group by voxel-based analysis. As the results, we identified the difference in brain volume change due to environment. In early developmental period (3-6-9 months), the volume of primary visual cortex (V1) was significantly lower in enriched marmosets than in standard group. In late developmental period (12-15-18 months), the volume difference in V1 was diminished. Instead, the volume of higher-order visual cortex was significantly decreased in enriched group. Our results demonstrated both standard and enriched group marmosets had similar physiques, which indicated the standard cage defined in this study had the enough size for physical development. On the other hand, environment had impact on brain development. The timing of rapid volume decrease in enriched marmosets would be earlier than that in standard marmosets in V1, suggesting enriched environment might accelerate brain maturation. These findings suggest rearing environment demonstrated neurodevelopment would be particularly sensitive to environment, and eventually had influence on modifying experimental result. Further studies about the influence of breeding environment on brain structure would contribute to provide more stable data in neuroscience research.

Disclosures: **F. Seki:** None. **T. Inoue:** None. **T. Yurimoto:** None. **M. Togashi:** None. **T. Muraoka:** None. **K. Mukasa:** None. **J. Hoshi:** None. **Y. Komaki:** None. **Y. Kurotaki:** None. **E. Sasaki:** None.

Poster

PSTR122. Animal Models of Behavioral Dysfunction

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR122.08/B37

Topic: A.09. Adolescent Development

Title: Age- and sex-dependent changes in cellular proliferation in the dorsal and ventral dentate gyrus

Authors: *R. D. ROMEO, N. GARCIA, A. RICHEY, C. PARKIN, J. ORTIZ, S. CRUZ; Barnard Col., New York, NY

Abstract: Adolescence is a stage of development associated with several major structural and functional changes in the brain. One key change is the significant reduction in cellular proliferation and neurogenesis in the dorsal dentate gyrus of the hippocampal formation, the physiological and behavioral implications of which are unclear. Lesion studies reveal that the dorsal hippocampus is involved in learning and spatial memory, whereas the ventral hippocampus regulates emotional and motivated behaviors. Although an adolescent-related decline in the number of immature neurons in the dentate gyrus has been established, relatively little is known about the changes in proliferation across the dorsal and ventral hippocampus and the role sex plays in these adolescent-related changes. To address this gap, we examined cellular proliferation using 5-bromo-2’deoxyuridine (BrdU) immunohistochemistry in 30 day old (pre-adolescent) and 70 day old (post-adolescent) male and female rats in both the dorsal and ventral dentate gyrus. Rats were injected with two doses of 200 mg/kg BrdU 24 hours apart at 28 and 29

(pre-adolescent) or 68 and 69 (post-adolescent) days of age. Rats were perfused 24 hours after the last BrdU injection (n = 6-8 per age group and sex). Statistical analyses revealed significant adolescent related decreases in cellular proliferation in the dorsal dentate gyrus in both males and females, with a more pronounced decrease in males. We found no age and sex-dependent changes in the proliferation of cells in the ventral dentate gyrus. These data indicate that adolescent changes in cellular proliferation are specific to the dorsal aspect of the dentate gyrus and suggest that the dorsal and ventral hippocampal formation might play different roles in cognitive and emotional changes observed during adolescence.

Disclosures: R.D. Romeo: None. **N. Garcia:** None. **A. Richey:** None. **C. Parkin:** None. **J. Ortiz:** None. **S. Cruz:** None.

Poster

PSTR122. Animal Models of Behavioral Dysfunction

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR122.09/Web Only

Topic: A.09. Adolescent Development

Title: Isolation Induced Anhedonic Behavior in Adolescent but not Adult Sprague-Dawley Rats

Authors: *R. BARNET, H. MCARDLE, S. WILLIAMS, T. ZAVAR;
Col. William & Mary, Williamsburg, VA

Abstract: The prevalence of mental illnesses such as depression and anxiety has risen drastically over the past few years suggesting long-lasting effects of the COVID-19 pandemic continue to be experienced. Studies using rodent models impose housing-related stressors such as social isolation and sensory-impoverished housing conditions suggest that these kinds of stressors are sufficient to induce depression-like behavior. The current research sought to explore this issue using the sucrose preference test, a rodent model of consumption measuring preference for sucrose compared to water in which a decrease in sucrose preference is taken to reflect anhedonic depression-like behavior. Interest was in age-dependent effects of social isolation and sensory-impoverished housing conditions versus social and environmental enrichment in rats on depressive anhedonia measured in the sucrose preference test, as well as anxiety measured in light-enhanced startle. In Experiment 1, housing manipulations were initiated in adulthood (approx. post-natal day 85) and in Experiment 2 in adolescence (approx. post-natal day 35). Sprague-Dawley male and female rats were singly housed in isolation, or in groups of 2-3 per cage with the addition of enhanced bedding, shelter, and ‘toys’ which rotated every 3 days. After 4 weeks and 11 weeks of housing exposure (Experiment 1) or after 4 weeks only (Experiment 2) subjects were tested in the sucrose preference test and light-enhanced startle (Experiment 2 only). Adult exposed rats failed to display any behavioral alteration as a result of housing condition following either 4 or 11 weeks of housing exposure. Adolescent exposed rats singly housed in a nonenriched environment showed a reduction in sucrose preference compared to group housed environmentally enriched counterparts following 4 weeks of housing

manipulation. Adolescent animals also displayed increased anxiety measured in the light-enhanced startle paradigm implicating alterations in the bed nucleus of the stria terminalis. These data suggest housing effects in rodents are age-dependent and isolation housing of adolescent but not adult rats induce an anhedonic depressive phenotype. Possible sex effects and critical variables in the sucrose preference test are discussed.

Disclosures: **R. Barnet:** None. **H. McArdle:** None. **S. Williams:** None. **T. Zavar:** None.

Poster

PSTR122. Animal Models of Behavioral Dysfunction

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR122.10/B38

Topic: A.09. Adolescent Development

Support: National Institute on Drug Abuse NIDA R01 DA037911
Canadian Institutes of Health Research FRN: 156272

Title: Social stress in adolescence induces sex-specific ectopic growth of mesolimbic dopamine axons to the prefrontal cortex

Authors: *S. RICHER[1,3], D. MACGOWAN[1,3], A. H. PANTOJA URBAN[1,3], A. SONG[1], S. GUL[1], G. A. HERNANDEZ[3], C. FLORES[3,2];
[2]department of Psychiatry, department of neurology and neurosurgery, [1]McGill Univ., Montreal, QC, Canada; [3]Douglas Mental Hlth. Univ. Inst., Montreal, QC, Canada

Abstract: Bullying in adolescence can hamper prefrontal cortex (PFC) development, impairing its role in controlling impulse behavior and negatively affecting psychopathology onset and severity. In adolescence, dopamine (DA) axons expressing the guidance cue receptor DCC undergo targeting events in the nucleus accumbens (NAcc), forming enduring synaptic connections. However, mesocortical DA axons continue to grow from the NAcc to the PFC over adolescence remaining vulnerable to experiences. Using an accelerated social defeat stress model for adolescent male mice (AcSD), we showed that social stress downregulates DCC receptors in DA neurons leading to altered PFC DA connectivity and inhibitory control deficits in adulthood. In this study, we tested if AcSD in adolescence induces the mistargeting of NAcc DA axons and their ectopic growth to the PFC in both male and female adult mice.To track adolescent PFC DA axon growth, we unilaterally microinjected a retrogradely transported virus expressing floxed Flp recombinase into the NAcc of postnatal day 21, DATCre male C57BL/6J mice. Simultaneously, we microinjected into the ipsilateral ventral tegmental area a Flp-dependent enhanced yellow fluorescent protein (eYFP) virus. Flp recombinase is therefore only expressed in Cre+ DA neurons that have reached the NAcc by early adolescence. At postnatal day 25, mice were exposed to AcSD or to control conditions. In adulthood, PFC DA axon growth was assessed by quantifying eYFP+ terminals with stereology.We find that male mice undergoing AcSD in adolescence show a greater number of eYFP+ axons in the adult PFC compared to controls.

Contrastingly, female mice undergoing AcSD show a decrease in eYFP+ axons growing to the PFC when compared to their control counterparts. This is the first demonstration that exposure to physical/psychosocial harm in adolescence can deviate DA axons from their intended target, inducing their input into off-target regions, and likely altering adult cognitive processing.

Disclosures: S. Richer: None. **D. MacGowan:** None. **A.H. Pantoja Urban:** None. **A. Song:** None. **S. Gul:** None. **G.A. Hernandez:** None. **C. Flores:** None.

Poster

PSTR122. Animal Models of Behavioral Dysfunction

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR122.11/B39

Topic: A.09. Adolescent Development

Title: Pubertal regulation of ventral tegmental area development

Authors: *V. RIESGO, T. BEEDY, K. THOMPSON, J. WILLING;
Psychology, Bowling Green State Univ., Bowling Green, OH

Abstract: The ventral tegmental area (VTA) contains dopamine-producing cells that are essential for behaviors such as reward processing, memory and cognition, sexual behavior, risk assessment, and mood regulation. Previous work has shown that dopamine cell number in the VTA changes between the juvenile and young adult period. Evidence also suggests that the VTA is sensitive to hormones like estrogen and testosterone, though a role for puberty in development of this region has been unexplored. Here, we determined the day of pubertal onset in male and female rats. Brain tissue was collected from pre-pubertal, 1 day post pubertal onset, 5 days post pubertal onset, and adult males and females. We immunohistochemically stained brain sections containing the VTA for tyrosine hydroxylase to assess the number of dopamine neurons in the region. Alternate sections were either nissl-stained or immunohistochemically stained for perineuronal nets (PNNs). Dopamine neurons, total neuron number and PNN number were stereologically quantified. We also measured the VTA optical density of TH using ImageJ software. We report sex-specific developmental changes in VTA neuroanatomy mediated by the onset of puberty. Results from these experiments have important implications for our understanding of the connection between dopamine, pubertal hormones, and synaptic plasticity during the adolescent period.

Disclosures: V. Riesgo: None. **T. Beedy:** None. **K. Thompson:** None. **J. Willing:** None.

Poster

PSTR122. Animal Models of Behavioral Dysfunction

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR122.12/B40

Topic: A.09. Adolescent Development

Support: NSERC 2018-03706
NSERC PGS-D
CFI 30215
CRC-2018-00023

Title: Adult prefrontal cortex neuroanatomy and social skills in rats reflect natural variation in juvenile play experiences

Authors: *J. HAM, A. N. IWANIUK, S. M. PELLIS;
Neurosci., Univ. of Lethbridge, Lethbridge, AB, Canada

Abstract: Juvenile social play is associated with the development of socio-cognitive skills and the neural mechanisms that control those skills. When juvenile play experiences are manipulated in rats, through social isolation or altered-peer associations, adult rats have difficulty inhibiting an escalation to aggression when interacting with strangers. Along with social deficits, the morphology of pyramidal neurons of the medial prefrontal cortex (mPFC) is altered; socially competent rats have smaller, pruned neurons than socially incompetent rats. Rats naturally exhibit individual variation in the style and frequency of juvenile play, but whether variation in social skills as adults is associated with variation in juvenile social play or the morphology of mPFC neurons remains unknown. In the present study, the social skills of 54 adult male rats, reared in groups of six, were assessed. When paired with an unfamiliar male in a neutral arena, 11/54 rats escalated to aggression, suggesting poor socio-cognitive skills. Comparing the 11 socially deficient rats to matched, socially competent group mates, two predictions were tested. First, the mPFC pyramidal neurons of the rats with poor social skills should have larger neurons with less dendritic pruning than rats that did not escalate to aggression. Second, the juvenile play experience of the rats with poor socio-cognitive skills should have been less or atypical play as juveniles. Using virtual microscopy to digitally reconstruct neuron morphology and highly refined measures of play (e.g., assessing asymmetry in the play of pairs of rats) these predictions were tested. As expected, a GLM revealed that the pyramidal neurons of rats with poor socio-cognitive skills had significantly less dendritic pruning (e.g., convex hull volume, $p < .0001$). In addition, even though socially incompetent juveniles played as much as their peers, their play was more aggressive, and they did not engage in group play like their peers (e.g., they did not engage in multi-animal play bouts). These results suggest that the variation in the quality of juvenile play experiences leads to changes in neuron morphology and ultimately affects adult social behavior. That is, the developmental benefits afforded by juvenile play, in male rats, are not gained equally among individuals.

Disclosures: J. Ham: None. **A.N. Iwaniuk:** None. **S.M. Pellis:** None.

Poster

PSTR122. Animal Models of Behavioral Dysfunction

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR122.13/B41

Topic: A.09. Adolescent Development

Support: MOE RG99/20

Title: Investigating the Role of Parent-Offspring Interactions on Early Social Learning Using Dyadic Optogenetics

Authors: *G. HAM[1], G. J. AUGUSTINE[1], V. LEONG[1,2];
[1]Nanyang Technological Univ., Singapore, Singapore; [2]Univ. of Cambridge, Cambridge, United Kingdom

Abstract: Although both rodents and humans possess the ability to learn vicariously through social interaction, rather than direct experience, the neural mechanisms that subserve social learning are poorly understood. Recent studies of human learning suggest that interbrain neural synchrony may be a potential mechanism that mediates social learning, possibly through interbrain coupling of neural oscillations (Trends in cognitive sciences. 2020. 24(4):329-342). In rodent models, optogenetic methods allow the precise control of neural activity that is required for testing of this hypothesis. Accordingly, here we report important progress toward the development of a novel parent-offspring (dyadic) optogenetic mouse model, which will permit synchronous and asynchronous optogenetic control of dam-pup neural activity to be achieved by P22. To test early social learning in rodents, we successfully adapted the (adult-adult) social transmission of food preference (STFP) paradigm for use with dam-pup dyads by age P24. Dams were fed one of two novel flavors and allowed to interact with their pups (age P24-30). Pups were subsequently exposed to the two flavors, with food consumption quantified as the learning outcome. Pups ate more of the flavor consumed by their mothers, indicating successful STFP from parent to offspring. Further, we developed a detailed behavior coding scheme to investigate moment-to-moment dynamics of dam-pup social interactions, integrating data streams from multiple camera views and ultrasonic microphones. We identified several candidate social behaviors relevant to STFP and found evidence suggesting a strong relationship between nose-to-nose interactions and learning outcomes ($R = 0.35$, $p = 0.05$). These results suggest that, similar to human interactions, face-to-face social interactions in rodents are crucial for the vicarious learning of food preferences from an early developmental stage. Finally, to allow artificial synchronization of dam-pup neural activity during learning, we have successfully implemented a wireless optogenetics system that allows pups as young as P16 to freely interact with their dams. This work paves the way for optogenetic experiments to access the contribution of dam-pup neural synchronization in different neural circuits toward social learning processes. Establishment of this rodent parent-offspring model represents a crucial first step toward understanding the neural mechanisms underlying social learning, with broader implications for the study of developmental disorders such as autism and learning disabilities, as well as disorders associated with deficits in parent-child engagement.

Disclosures: G. Ham: None. **G.J. Augustine:** None. **V. Leong:** None.

Poster

PSTR122. Animal Models of Behavioral Dysfunction

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR122.14/B42

Topic: A.09. Adolescent Development

Support: NRF-2020R1A2C2012416
IBS-R015-D1

Title: Social isolation reduces functional segregation of sensory networks while enrichment enhances multimodal areas from sensory stimulation

Authors: *T. YOU[1,2], T. LEE[3], G. IM[4], M. JANG[5], S.-G. KIM[1,2], S. CHUNG[6], J. LEE[5,2,4]; [1]Ctr. For Neurosci. Imaging Res., Suwon, Korea, Republic of; [2]Biomed. Engin., Sungkyunkwan Univ., Suwon, Korea, Republic of; [3]Korea Brain Res. Inst., Daegu, Korea, Republic of; [4]Ctr. for Neurosci. Imaging Res., Suwon, Korea, Republic of; [5]Radiology, Sungkyunkwan Univ. Sch. of Med., Seoul, Korea, Republic of; [6]Physiol., Sungkyunkwan Univ. Sch. of Med., Suwon, Korea, Republic of

Abstract: Environmental factors play an important role in the structural and functional development of the brain. Proper brain function and development relies on the maturation of unique brain systems that results in specialized networks. The effects of social isolation or environmental enrichment are well studied on the neurochemical, cognitive, anatomical, and local functional level, but how they affect the global brain network is not fully understood. In this study, we housed P28 mice in either social isolation (SI) (n = 8), standard cage (n = 12), or enriched environment (EE) (n = 12) for 6 weeks. We utilized functional magnetic resonance imaging (fMRI) with a 15.2 Tesla magnet to acquire brain wide functional response (TR/TE = 1000/11.5 ms, 0.132 x 0.132 x 0.5 mm resolution, 18 slices) from four sensory stimulations: whisker-pad, forepaw, visual, and olfactory. We also characterized the brain's basal network with resting state functional connectivity (RSFC) analysis to calculate basal and evoked system segregation. We found that socially isolated mice have reduced segregation between the whisker/forelimb and orofacial network to the posterior lateral network (PLN), which composes of visual and auditory areas. When visually stimulated, SI mice had reduced response within their PLN while response was elevated in the whisker/forelimb and orofacial networks. This suggests SI results in a loss of specialization of the PLN's visual function and spreads the functionality to the somatosensory areas. In addition, SI mice had elevated response, particularly in hypothalamus and limbic regions, to olfactory stimulation. Interestingly, both SI and EE group had elevated response in the retrosplenial cortex and anteromedial visual area from whisker or forepaw stimulation suggesting these areas are key integration regions. Last, EE group had enhanced response in sensorimotor areas such as ventral posterior, posterior, and ventral-anterolateral thalamus, caudate putamen, and midbrain reticular nucleus suggesting enrichment enhances sensorimotor function. Taken together, we show sensory network remapping due to social isolation which results in reduced system segregation and alters sensory stimulation response. Our results suggest social isolation mimics network changes that are similar to

depression and schizophrenia. Interestingly, enrichment did not alter system segregation, but had enhanced response in multimodal areas suggesting improved sensory integration function.

Disclosures: **T. You:** None. **T. Lee:** None. **G. Im:** None. **M. Jang:** None. **S. Kim:** None. **S. Chung:** None. **J. Lee:** None.

Poster

PSTR122. Animal Models of Behavioral Dysfunction

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR122.15/B43

Topic: A.09. Adolescent Development

Support: FAPERJ E-26/201.081/2022
FAPERJ E26/010.002428/2019
CNPq 313353/2021-2
CAPES 001

Title: Neurochemical and behavioral effects of varenicline treatment in a mice model of tobacco-smoke exposure during adolescence

Authors: *A. C. MANHAES, C. K. G. BRANDÃO, F. UCHOA-BRAGA, J. ISNARDO-FERNANDES, A. NUNES-FREITAS, R. EFRAIM-CORREA, A. M. DO NASCIMENTO, A. RIBEIRO-CARVALHO, C. C. FILGUEIRAS, Y. ABREU-VILLAÇA;
Physiological Sci., Univ. Estado do Rio de Janeiro, Rio de Janeiro, Brazil

Abstract: The use of tobacco products during adolescence generates nicotine dependence quickly and there is greater difficulty in maintaining abstemiousness when trying to stop use. Varenicline (VAR), a partial α4β2 and total α7 nicotinic cholinergic receptor agonist, is one of the drugs approved for the treatment of nicotine-associated substance use disorder in adults. This substance was used here in the treatment of an animal model of exposure to cigarette smoke during adolescence with the aim of evaluating its effects on neurochemical and behavioral parameters. From postnatal day 30 (PN30) to 45, 224 Swiss mice were exposed to cigarette smoke (0.73 mg nicotine; SMK) or air (AIR) for 8 h/day in automated exposure. At PN41, treatment with VAR (0.5 mg/kg/bid v.o.) or water (WTR) was initiated; from PN46 to PN56, the dose was doubled. Part of the animals was euthanized at PN41 for neurochemicals analyzes (n=8 per group). The other part was tested (n=24 per group) in the Elevated Plus Maze (EPM) and in Hole Board (HB) during the treatment period (PN54/PN55), before being euthanized at PN56 for neurochemical analysis. SMK exposure for eleven days (PN41) did not affect alfa-4, alfa-7 and beta-2 subunits of cholinergic receptors, the contents of the tyrosine hydroxylase enzyme (TH) and of the dopamine transporter protein (DAT) in the hippocampus. At PN56, at the end treatment, increased levels of the alfa-4, but not of beta-2, in adolescent females were detected. VAR down-regulated alfa-7 in SMK animals. Exposure to SMK followed by VAR altered DAT levels in males but not of females. SMK males showed an increase in behaviors associated with

anxiety in the EPM when compared to AIR VAR normalized male locomotor activity, which had been reduced by SMK. In the HB, SMK animals showed a reduction in the behavior associated with novelty seeking when compared to AIR ones. In females, VAR normalized the increase in anxiety-like behaviors caused by SMK. Our results indicate that, although the use of VAR is generally considered to be safe, this drug has a sex-dependent effect on the dopamine neurochemical circuitry in adolescent animals. These findings indicate that the study of the effects of the use of VAR in the treatment of adolescent smokers should be deepened.

Disclosures: **A.C. Manhaes:** None. **C.K.G. Brandão:** None. **F. Uchoa-Braga:** None. **J. Isnardo-Fernandes:** None. **A. Nunes-Freitas:** None. **R. Efraim-Correa:** None. **A.M. do Nascimento:** None. **A. Ribeiro-Carvalho:** None. **C.C. Filgueiras:** None. **Y. Abreu-Villaça:** None.

Poster

PSTR122. Animal Models of Behavioral Dysfunction

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR122.16/B44

Topic: A.09. Adolescent Development

Title: Chronic stress during development affects the epididymal oxidant state in rats

Authors: *Y. DE LEÓN RAMÍREZ[1,2], V. NOLASCO-ORDOÑEZ[3], X. HERNÁNDEZ-DOMÍNGUEZ[4], R. COUTIÑO-RODRÍGUEZ[2], L. NICOLAS TOLEDO[3], O. ARROYO-HELGUERA[2];
[1]Inst. de Salud Pública, UV, Tlaxcala, Mexico; [2]Inst. de Salud Pública, UV, Xalapa, Mexico; [3]Ctr. Tlaxcala de Biologia de la Conducta, Univ. Autonoma de Tlaxcala, Tlaxcala, Mexico; [4]Univ. Veracruzana, Xalapa, Mexico

Abstract: Stressful events exposure at early age induces changes in the hypothalamus-pituitary axis that results in glucocorticoid chronic secretion, which in turn affects spermatogenesis in adulthood. Moreover, spermatozoa maturation depends upon the oxidation state within epididymal tissue whose maturation and storage role are fundamental in spermatogenesis, as sperm apoptosis and loss of motility was described when the epididymal oxidant state is altered, at least in adult rats. However, few studies during the development have been carried out taking into consideration oxidant state and/or sperm quality. Here, we assessed sperm viability, motility and concentration, and oxidant state of young rats after chronic stress induced by space reduction, restraint, and forced swimming. We used a control and experimental group (n=8), the experimental group underwent a one-a-day alternate stressful trial on a daily basis for 4 weeks, these trials included: space reduction placing 8 rats in a small cage during 5 h; forced swimming during 10 min; and placing in a restrictor during 3h. Chronically stressed animals presented low sperm concentration, decreased sperm motility, and viability, likewise, oxidative stress markers like TBARS, FRP, and Cas3 were higher in experimental groups; although SOD, CAT, and GSHt were similar to control group values. Present results suggest that chronic stress stimuli

during puberty in young rats have detrimental effects on sperm quality which may contribute to fertility issues later in life. It is likely that nociceptors, proprioceptors, and chemoreceptors sensory information spinothalamic dorsal-ventral tracts reach the UPL nucleus from here to several nuclear within the hypothalamus. Traveling through ventral and dorsal spinothalamic tracts reaches the ventral posterior lateral is within the thalamus and from here to several nuclei hypothalamus which activates the hypothalamus-pituitary axis inducing a neuroendocrine reflex response of glucocorticoids release.

Disclosures: **Y. De León Ramírez:** None. **V. Nolasco-Ordoñez:** None. **X. Hernández-Domínguez:** None. **R. Coutiño-Rodríguez:** None. **L. Nicolas Toledo:** None. **O. Arroyo-Helguera:** None.

Poster

PSTR122. Animal Models of Behavioral Dysfunction

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR122.17/B45

Topic: A.09. Adolescent Development

Support: R01-MH086507
R01-MH127850

Title: Developmental disruption of mediodorsal thalamus-prefrontal pathway by early life adversity

Authors: *D. NEAL, K. Y. TSENG;
Anat. and Cell Biol., Univ. of Illinois Chicago, Chicago, IL

Abstract: Exposure to adversity early (ELA) often leads to detrimental cognitive phenotypes that yield deficits in threat processing, working memory, and social behavior. Accordingly, research has been focused on the developmental impact of ELA on key corticolimbic structures including the prefrontal cortex (PFC), hippocampus, and amygdala. Yet, proper functioning of the PFC and associated circuits requires the integration of inputs from the mediodorsal thalamus (MD), which is also involved in the regulation of cognitive functions. Here we asked whether the MD-to-PFC pathway is developmentally regulated and susceptible to disruptions during a critical period by maternal separation ELA. Using *in vivo* electrophysiology, we found that the pattern of PFC local field potential responses to MD train stimulation is developmentally regulated through adolescence in both males and female rats. Typically, a local field potential facilitation emerges in the adult PFC at 10Hz while a suppression of the response occurs at 20Hz and 40Hz. Following maternal separation ELA from P11-20, a similar pattern of local field potential response was observed in adulthood. However, the magnitude of PFC facilitation and suppression was markedly reduced in ELA rats, suggesting that both excitatory and inhibitory mechanisms of MD-evoked transmission are compromised. Ongoing studies will test the impact

of maternal separation ELA from P2 to P10 to reveal if there is a critical developmental window of susceptibility to ELA.

Disclosures: D. Neal: None. **K.Y. Tseng:** None.

Poster

PSTR122. Animal Models of Behavioral Dysfunction

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR122.18/B46

Topic: A.09. Adolescent Development

Title: Prior experience does and does not alter evoked activity in medial entorhinal cortex of adolescent rats exploring a space with or without objects

Authors: *E. N. SHEFFIELD, B. M. HAYES, A. L. KELLEY, M. C. ZRULL;
Psychology, Appalachian State Univ., Boone, NC

Abstract: The medial entorhinal cortex (MEC) leads, and ends, a neural processing loop through the hippocampal formation (HF). In part, this loop contributes to neural representations of an animal's environment as a whole that is updated as an animal interacts with the space. We used c-FOS to examine neural activation in Layers 2 and 3, which receive input from various brain regions and relay input to the HF, and Layer 5 of MEC, which receives HF output and is reciprocally connected with Layers 2 and 3, evoked by exposure to a new or known, multi-level environment with or without a variety of moveable objects. After weaning, 37 rats were group housed in standard shoebox cages and spent 1.5 h, 5 days/week in an enclosure with ramps and platforms with (EEob) or without (EEno) objects or were only handled (No group). At 49 days old and prior to sacrifice, rats from each experience group spent 1.5 h in the enclosure with (No-EEob, EEob-EEob, EEno-EEob groups) or without (No-EEno, EEob-EEno, EEno-EEno) or in a standard cage in a quiet and dark room (controls⋯ No-No, EEobNo, EEnoNo). Brains were processed to visualize c-FOS+ neurons, and cell counts were made using digital microscopy and stereological technique. Data were standardized to No-No group cell counts. For Layers 2 and 3 of MEC, any exposure to the environment, whether with or without objects and without regard to prior experience in the setting, increased neural activity over no exposure (No-No controls), +68% ($p<.001$) and +84% ($p=.015$), respectively. These data suggest activity in superficial MEC is dependent upon the current situation without significant regard to prior experience in a similar or different setting (i.e. with or without objects). In contrast, Layer 5 evoked activity depended upon prior experience ($p<.001$). Neural activity did increase in rats with no prior experience in the environment whether objects were present or not (+150% for both No-EEob and No-EEno over No-No). For rats with a history of sessions in the environment, increases in neural activity above controls were greatest when the historical space did not match the current space. Specifically, active neurons for EEob-EEno were +242% and for EEob-EEob +138% over EEob-No, and were +285% for EEno-EEob and 175% for EEno-EEno over EEno-No. The data suggest evoked neural activity in MEC Layer 5 was dependent upon both prior experience and the

current situation. Greater numbers of active Layer 5 neurons in response to an environment that did not exactly match a previously experienced space supports the idea that MEC provides a representation of the environment dependent, at least to some degree, upon interaction with the space.

Disclosures: **E.N. Sheffield:** None. **B.M. Hayes:** None. **A.L. Kelley:** None. **M.C. Zrull:** None.

Poster

PSTR122. Animal Models of Behavioral Dysfunction

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR122.19/B47

Topic: A.09. Adolescent Development

Support: National Institute on Drug Abuse NIDA R01 DA037911
Canadian Institutes of Health Research FRN: 156272

Title: Non-invasive miR-218 downregulation as a potential therapeutic approach to mitigate long-term effect of amphetamine during adolescence.

Authors: *M. TEIXEIRA[1], *M. TEIXEIRA[1], *M. TEIXEIRA[1], R. AVRAMESCU[1], A. TORRES-BERRIO[3], A. SONG[4], G. HERNANDEZ[4], C. FLORES[2,4];
[1]Integrated Program in Neurosci., [2]Psychiatry, Neurol. and Neurosurg., McGill Univ., Montreal, QC, Canada; [3]Lurie Ctr. for Autism, Harvard Med. Sch., Boston, MA; [4]Douglas Mental Hlth. Univ. Inst., Montreal, QC, Canada

Abstract: Adolescence is a critical developmental period for decision-making and behavioral inhibition, coinciding with the protracted maturation of the mesocorticolimbic dopaminergic pathway. The Netrin-1/DCC axonal guidance cue system is critically involved in this process. Furthermore, recreational-like doses of amphetamine in adolescent male mice downregulate DCC receptors in dopamine neurons, by recruiting the microRNA miR-218, which is potent repressor of this transcript. This process results in ectopic growth of mesolimbic dopaminergic axons to the prefrontal cortex (PFC), leading to long-term deficit in impulse control. In this study, we investigated the potential of noninvasive nasal administration of an anti-miR oligonucleotide to downregulate miR-218, thereby mitigating the deleterious long-term effects of amphetamine treatment during adolescence. To achieve this, we employed a locked nucleic acid (LNA) oligonucleotide targeting miR-218 (anti-miR-218). First, we validated the distribution of the anti-miR-218 within the brain by intranasal administration (n=4) of a modified Cy3-tagged version using a dose of 5 µl of 0.3 mM. Brain perfusion was performed 48 hours post-administration and presence of the molecule was investigated using confocal microscopy. Anti-miR-218 was detected in various brain regions, including the olfactory bulb, the PFC, and the ventral tegmental area (VTA). Subsequently, using the untagged version of ant-miR-218 (n=8) and a scrambled LNA sequence as control (n=7), we measured miR-218 brain expression with RT-qPCR analysis. We demonstrated that a substantial amount of anti-miR-218 reaches the

brain, enough to significantly downregulate miR-218 by 48h in several different brain regions including PFC, nucleus accumbens and VTA. Our findings suggest that intranasal anti-miR-218 effectively reaches different brain regions, and successfully downregulates miR-218 expression within 48 hours, representing a promising noninvasive approach utilizing a LNA oligonucleotide to downregulate miR-218 levels in the brain. With this approach, we aim to counteract the detrimental effects of amphetamine treatment on dopaminergic pathway maturation. Our study is timely as it serves to address the rise in stimulant consumption among youth in North America, according to the Inter-American Drug Abuse Control Commission report of 2022.

Disclosures: **M. Teixeira:** None. **M. Teixeira:** None. **M. Teixeira:** None. **R. Avramescu:** None. **A. Torres-Berrio:** None. **A. Song:** None. **G. Hernandez:** None. **C. Flores:** None.

Poster

PSTR122. Animal Models of Behavioral Dysfunction

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR122.20/B48

Topic: A.09. Adolescent Development

Support: NIH Grant DA056288
NIH Grant DA013137
NIH Grant DA059310
NIH Grant NS100624

Title: Adolescent Oral Oxycodone Self-Administration Alters Neurobehavioral and Neurocognitive Development

Authors: *K. A. MCLAURIN, C. F. MACTUTUS, R. M. BOOZE;
Psychology, Univ. of South Carolina, Columbia, SC

Abstract: The adolescent-associated shift in the relative balance of dopaminergic system function renders adolescents and young adults uniquely vulnerable to substance use initiation. Indeed, nonmedical use of prescription opioids peaks during late adolescence, whereby approximately 2.8% of adolescents and 5.6% of young adults develop opioid use disorder (OUD). Although generalized neurocognitive dysfunction is recognized as one of the devastating consequences of OUD in adults, extrapolating cross-sectional findings to developmental processes is inferentially fraught. Hence, the rationale was built for utilizing a longitudinal experimental design to establish the dose-dependency of adolescent OXY self-administration on the trajectory of neurobehavioral and neurocognitive development. From postnatal day (PD) 35 to PD 105, an age in rats that corresponds to the adolescent and young adult period in humans, male and female F344/N rats (*n*=12 rats per group) received access to either oral OXY (0, 5, or 10 mg/kg) or water under a two-bottle choice experimental paradigm. Repeated assessments (i.e., every 30 days from PD 30 to PD 180) of locomotor activity and prepulse inhibition were conducted to examine OXY induced behavioral (e.g., hyperactivity) and neurocognitive (e.g.,

temporal processing) impairments. Animals exhibited a voluntary escalation of OXY intake across time, whereby the rate of escalation was significantly faster in female animals relative to their male counterparts. Examination of gross-motoric system function during a 60-minute locomotor activity test session revealed dose-dependent hyperactivity; hyperactivity that was independent of biological sex and persisted despite abstinence from OXY. Furthermore, dose-dependent hyperreactivity and temporal processing impairments were evidenced by developmental alterations in visual prepulse inhibition. Fundamentally, at PD 180, animals with a history of adolescent OXY self-administration exhibited pronounced deficits in prepulse inhibition. Taken together, adolescent oral OXY self-administration induces long-term alterations in neurobehavioral and neurocognitive development necessitating the implementation of safer prescribing guidelines for this population. Funded by: DA056288, DA013137, DA059310, NS100624.

Disclosures: K.A. McLaurin: None. **C.F. Mactutus:** None. **R.M. Booze:** None.

Poster

PSTR122. Animal Models of Behavioral Dysfunction

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR122.21/B49

Topic: A.09. Adolescent Development

Title: Brain functional connectivity and the influence of an enriched enviroment in rats from childhood to adolescence

Authors: *E. GONZÁLEZ-PÉREZ[1], J. ORTIZ-RETANA[1], J. ALFARO-MORENO[1], S. ALCAUTER[2];
[1]Inst. de Neurobiologia, UNAM, Queretaro, Mexico; [2]Neurobiología Conductual y Cognitiva, Inst. De Neurobiologia. Univ. Nacional A, Queretaro, Mexico

Abstract: Enriched environment (EE) refers to a condition that offers increased opportunities for exploration and stimulation. It has been found to enhance cognitive and motor skills in animals. For instance, mice in an enriched environment showed improved performance in spatial memory tasks compared to those in standard environments. The study of structural and functional properties of animal brains often involves techniques such as magnetic resonance imaging (MRI), which allows for the measurement of functional connectivity (FC) - the temporal correlation between anatomically separated brain regions. In this study, we examined the development of the default mode network (DMN) and sensorimotor network (SMN) in rats from childhood to adolescence, as well as their performance in a spatial memory task after exposure to either an enriched environment or a standard environment. A total of 24 rats (EE=14, control=10) aged 21 and 45 postnatal days were included in the study. The control group was housed in standard cages, while the EE group had access to larger cages equipped with a training wheel, tunnels, and toys. Functional connectivity between the regions of the DMN (Retrosplenial cortex and Cingulum) and the SMN (motor cortices left and right) was assessed using a 7 Tesla

MRI scanner. The results showed a significant effect of age on the FC of the DMN, with increased FC observed in the EE group at postnatal day 45 (p=0.021). Additionally, the EE group exhibited better performance in the water maze task as the number of sessions increased. However, at postnatal day 21, there was a significant difference in performance throughout the sessions (p=0.04). In conclusion, exposure to an enriched environment has positive effects on learning, spatial memory, and functional connectivity within the DMN and SMN. Rats living in an enriched environment demonstrated higher connectivity and superior performance in the water maze task compared to control rats at postnatal day 45.

Disclosures: E. González-Pérez: None. **J. Ortiz-Retana:** None. **J. Alfaro-Moreno:** None. **S. Alcauter:** None.

Poster

PSTR122. Animal Models of Behavioral Dysfunction

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR122.22/B50

Topic: A.09. Adolescent Development

Support: NIDA R01DA046794

Title: Co-administration of alprazolam and fluoxetine during adolescence increases anxiety-like behavior and enhances drug reward sensitivity to fentanyl in male mice.

Authors: *N. I. VARDELEON[1], A. M. CARDONA-ACOSTA[2], N. MEISSER[2], A. BOWRING[2], K. J. BRISENO[2], C. A. BOLAÑOS-GUZMÁN[2];
[1]Psychological and Brain Sci., [2]Texas A&M Univ., College Station, TX

Abstract: CO-ADMINISTRATION OF ALPRAZOLAM AND FLUOXETINE DURING ADOLESCENCE INCREASES ANXIETY-LIKE BEHAVIOR AND ENHANCES DRUG REWARD SENSITIVITY TO FENTANYL IN MALE MICE.
NI. Vardeleon; AM. Cardona-Acosta; N. Meisser; A. Bowring; J. Briseno; CA. Bolaños-GuzmánTexas A&M University, (nvardeleon@tamu.edu)
According to the US National Comorbidity Survey Replication, there is strong correlation between major depressive and anxiety disorders in the general population. Unfortunately, the prevalence of multiple comorbidities is projected to significantly increase and emerge at even younger ages over the coming decades. Selective Serotonin Reuptake Inhibitors (SSRIs) such as fluoxetine (FLX; Prozac), are the first line of pharmaceuticals to treat adolescent depression and are often co-prescribed benzodiazepines such as alprazolam (ALP; Xanax) when anxiety symptoms are co-present. Evidence indicates that prolonged FLX exposure during adolescence results in increased sensitivity to anxiety-eliciting environments along with increased valence for both natural and drug reward. Recently, we have demonstrated that prolonged treatment with ALP during adolescence can potentiate reward valence for subthreshold (i.e., non-rewarding) doses of morphine (MOR) in adolescent male mice. The present study therefore was designed to

investigate whether short (7 days) co-treatment of ALP and FLX could influence/potentiate opioid reward in adolescent male mice. Postnatal day [PD] 35 C57BL/6 mice were treated with either a vehicle (VEH), ALP (0.5 mg/kg), FLX (20 mg/kg), or ALP+FLX (0.05 + 20 mg/kg) twice a day for 7 days (PD35-41). Twenty-four hours after the last injection, the mice were then subjected to the elevated plus-maze and the open field arena to assess for anxiety-like behavior(s). In addition, the mice were tested using the conditioned place preference (CPP) paradigm, a behavioral assay designed to assess drug reward. Here, we report that mice pretreated with FLX alone and ALP+FLX spent significantly less time in the center of the OFT indicating an increase in anxiety-like behavior(s). These results were not due to drug-induced changes in locomotor activity as there were no differences in distance traveled between the treatment groups. When tested for drug reward sensitivity to subthreshold (0.0125 mg/kg) or rewarding (0.025, 0.05 mg/kg) doses of fentanyl (FENT), ALP-, FLX-, and ALP+FLX-exposed mice spent significantly more time in the CPP compartments paired with the lowest dose of FENT. Together, our results suggest that co-treatment of FLX+ALP increases anxiety-like behavior and increases sensitivity to the rewarding properties of opioids such as FENT.

Disclosures: **N.I. Vardeleon:** None. **A.M. Cardona-Acosta:** None. **N. Meisser:** None. **A. Bowring:** None. **K.J. Briseno:** None. **C.A. Bolaños-Guzmán:** None.

Poster

PSTR122. Animal Models of Behavioral Dysfunction

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR122.23/B51

Topic: A.09. Adolescent Development

Support: RO1MH110921
RO1 MH075916
P50MH096891

Title: Mice treated with risperidone during the juvenile period sustain long term changes in social memory and PFC neuronal activity in adulthood

Authors: *C.-G. HAHN[1], W. ZHANG[2], A. WU[2], M. MSACKYI[3], K. BORGMANN-WINTER[2];
[1]Thomas Jefferson Univ., Bryn Mawr, PA; [2]Thomas Jefferson Univ., Philiadiaphia, PA; [3]Thomas Jefferson Univ., Philadelphia, PA

Abstract: Adolescence is a crucial stage for the development of exploratory behaviors and cognitive control functions. Over the last decade, prescription of antipsychotics for off-label indications in adolescence has drastically increased, affecting almost 2% of persons under 18. Moreover, a higher proportion of young male patients are administered antipsychotic medications for off-label use and for longer durations of time. Given the vulnerability of the developing brain, a pressing concern is off-target CNS effects of these agents which may be

sustained beyond the treatment period. Neurobiological effects of antipsychotics have been extensively studied but mostly in adulthood, but not in adolescence. During this highly dynamic period of brain development, we asked how antipsychotics affect exploratory and cognitive behaviors and brain circuits acutely and chronically. In this study, we treated WT mice with IP injection of risperidone (4mg/kg) (n=16) or saline (n=14) daily from P28-P49 (21 days). During adulthood (P62-P110), we tested the effect of risperidone on working memory, social memory, anxiety levels and spatial memory using Y-maze, three chamber social interaction, open field and novel object recognition. We found that the mice that were treated with risperidone during the juvenile period showed decreased social recognition and increased anxiety compared to those treated with the vehicle in the adulthood. Notably, male mice showed greater impairment compared to females in social recognition and open field test. We then examined these mice by employing *in vivo* Ca^{2+} imaging using two-photon microscope focusing on layer 2 neurons of the M2 cortex of awake animals during adulthood. Mice treated with risperidone during the juvenile period demonstrated decreased frequency in neuron firing compared to controls. Taken together, our results suggest that adolescent risperidone treatment may induce long-term changes in social memory and anxiety levels, associated with altered neuronal activity in the frontal cortex. Our results, while preliminary, may offer additional consideration in off-label treatment of adolescents with antipsychotics and suggest a possible relevance of sex differences in these effects.

Disclosures: C. Hahn: None. **W. Zhang:** None. **A. Wu:** None. **M. Msackyi:** None. **K. Borgmann-Winter:** None.

Poster

PSTR122. Animal Models of Behavioral Dysfunction

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR122.24/B52

Topic: A.09. Adolescent Development

Support: NSERC
CFI

Title: Exposure to stress during adolescences alters safety learning and emotional behaviours that persists into adulthood

Authors: G. SILVER, H. LEHMANN, *N. FOURNIER;
Psychology, Trent Univ., Peterborough, ON, Canada

Abstract: The ability to differentiate between signals that predict threat from those associated with safety is crucial for survival. Failure to properly utilize environmental cues that should signal “safety” from threats can lead to enhanced and maladaptive fear responses that are often seen in anxiety disorders. Adolescence is a critical period of transition that is marked by significant changes involving limbic and cortical regions important in social, cognitive, and

affective behaviours. Stressful life experiences during this period are known risk factors for the development of several neuropsychiatric conditions in adulthood, including anxiety, depression, posttraumatic stress disorder and substance abuse. However, the specific impact of adolescent stress on the ability of adults to utilize safety learning to inhibit fear responses remains poorly understood. To address this gap in knowledge, our study aimed to examine the impact of intermittent physical stress during the mid-adolescent period (PND 31-43) on safety learning, anxiety- and depressive-like behaviours in adult male Sprague Dawley rats (PND 70-91). Our findings revealed that exposure to mid-adolescent stress resulted in higher levels of defensive freezing along with reduced freezing responses to safety signals during a retention test of safety learning in adult rats. Additionally, these rats displayed increased anxiety-like behavior, as indicated by reduced exploration of the open arms in the elevated plus maze, and higher levels of immobility in the forced swim test, a measure of depression-related behavior. Finally, we observed a significant reduction in the number of reelin-positive cells, an important extracellular glycoprotein involved in synaptic plasticity and neurodevelopmental processes, in the medial prefrontal cortex of rats exposed to adolescent stress. In summary, our findings demonstrate that exposure to stress during critical periods of adolescence can have lasting effects on anxiety- and depressive-like behaviors, as well as impairments in utilizing safety signals to inhibit fear responses. Moreover, the observed reduction in reelin-positive cells suggests potential neurobiological mechanisms underlying these behavioral changes. These results also underscore the vulnerability of specific periods during adolescent development that are sensitive to the effects of stress, which can have enduring consequences on brain development, behaviour, and mental health.

Disclosures: **G. Silver:** None. **H. Lehmann:** None. **N. Fournier:** None.

Poster

PSTR122. Animal Models of Behavioral Dysfunction

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR122.25/B53

Topic: A.09. Adolescent Development

Support: The Natural Sciences and Engineering Research Council of Canada (NSERC)
The Canadian Institutes of Health Research (CIHR)
Canada First Research Excellence Fund (CFREF) for BrainsCAN at Western

Title: Neuronal and molecular maladaptation produced by adolescent edible delta-9-tetrahydrocannabinol (THC) consumption.

Authors: ***M. DEVUONO**[1,4], S. M. ANDERSON[1,4], M. PUSPARAJAH[2], J. GALINDO LAZO[2], M. H. SARIKAHYA[1,4], M. DE FELICE[1,4], H. J. SZKUDLAREK[1,4], K. K.-C. YEUNG[2], S. R. LAVIOLETTE[1,3,4];

[1]Anat. and Cell Biol., [2]Biochem. and Chem., [3]Psychiatry, Univ. of Western Ontario, London, ON, Canada; [4]Lawson Hlth. Res. Inst., London, ON, Canada

Abstract: Adolescent cannabis use is associated with an increased risk of neuropsychiatric illness later in life, yet rates of use in the age group remain high. Exposure to the psychoactive component of cannabis, Δ9-tetrahydrocannabinol (THC), during this critical period of brain development, can disrupt normal endocannabinoid system function, leading to long-term consequences. We have previously shown in rats that adolescent oral consumption of THC edibles, a popular method of cannabis use in humans, produces sex-dependent effects on anxiety and cognition, resulting in anxiety behaviour in males and cognitive deficits in both males and females. However, the underlying mechanisms remain undetermined. Previous evidence suggests that disturbances in mesocorticolimbic circuits (i.e., prefrontal cortex [PFC], ventral tegmental area [VTA] and nucleus accumbens [NAc]) may contribute to the long-lasting behavioural abnormalities induced by adolescent THC exposure. The current project explores the impact of adolescent oral THC consumption on mesocorticolimbic neuronal activity and neurotransmitter levels to gain insight into the mechanisms underlying the sex-specific behavioural changes. Adolescent male and female Sprague Dawley rats were given edibles containing increasing doses of THC (1-5 mg/kg) in Nutella® twice daily from postnatal day 35-45. In adulthood, i*n vivo* electrophysiology was then used to determine changes in PFC glutamatergic and VTA dopaminergic activity. Local field potentials in the PFC were also analyzed. Additionally, matrix-assisted laser desorption/ionization imaging mass spectrometry (MALDI IMS) was used to image the distribution of glutamate, GABA and monoamine neurotransmitters in PFC-NAc-VTA pathways. Overall, edible THC consumption during adolescence produced sex-dependent neuronal and molecular alterations which persist into adulthood. Adolescent edible THC resulted in PFC hyperactivity in males and females. THC also increased VTA dopaminergic bursting activity in males but decreased it in females. THC in females also led to a broad reduction in PFC oscillatory power, whereas only theta frequency power was reduced in males. MALDI IMS revealed profound long-term alterations of glutamate and GABA in the PFC and NAc of both males and females. Despite the sex differences in the effect on VTA bursting activity, THC reduced NAc dopamine in both sexes. Adolescent THC reduced norepinephrine (NE) exclusively in the PFC and NAc of females. The differences in the effect on oscillatory patterns, VTA activity, and NE may mediate the behavioural sex differences induced by adolescent edible THC.

Disclosures: M. Devuono: None. **S.M. Anderson:** None. **M. Pusparajah:** None. **J. Galindo Lazo:** None. **M.H. Sarikahya:** None. **M. De Felice:** None. **H.J. Szkudlarek:** None. **K.K. Yeung:** None. **S.R. Laviolette:** None.

Poster

PSTR122. Animal Models of Behavioral Dysfunction

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR122.26/B54

Topic: A.09. Adolescent Development

Support: NIH Grant OD011132
NIH Grant MH117103
HHMI Gilliam Fellowship

Title: Isolation in adolescence imperils social value-based decision-making later in life

Authors: *Y. GARCIA-SIFUENTES[1], E. SEO[2], A. ALLEN[2], L. BUTKOVICH[2], H. GIL-HENN[3], S. GOURLEY[2];
[1]Pediatrics, [2]Emory Univ., Atlanta, GA; [3]The Azrieli Fac. of Med., Bar-Ilan Univ., Safed, Israel

Abstract: Social isolation during adolescence leads to lasting behavioral consequences. However, how developmental isolation impacts decision-making behavior influenced by social experiences has yet to be comprehensively examined, and underlying mechanistic factors remain unclear. Here, mice were isolated from postnatal day (P) 31-56 and then reintegrated into social groups. We then trained mice to nose poke in operant conditioning chambers for two equally-preferred food reinforcers. Then, one food was paired with a novel conspecific, and the other with a novel object. Control mice later responded more for the conspecific-associated food. This preference was ablated in mice that had experienced adolescent isolation, and not isolation before or after this period. Next, we found that adolescent isolation elevates levels of the stress-sensitive Proline-rich tyrosine kinase 2 (Pyk2) in the basolateral amygdala (BLA). Further, Pyk2 overexpression in socially reared mice ablated social incentivization of future choice - recapitulating the effects of isolation. Thus, social isolation during adolescence obscures the ability of social experience to incentivize instrumental choice behavior, and these failures in adaptive choice persist despite normalization of the social milieu. Further, Pyk2 may be a mechanism by which social isolation during adolescence derails the ability of social experience to incentivize choice behavior later in life.

Disclosures: Y. Garcia-Sifuentes: None. **E. Seo:** None. **A. Allen:** None. **L. Butkovich:** None. **H. Gil-Henn:** None. **S. Gourley:** None.

Poster

PSTR122. Animal Models of Behavioral Dysfunction

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR122.27/B55

Topic: A.09. Adolescent Development

Title: Postnatal maturation of social behaviors in mice requires the brain specific transcription factor Npas4.

Authors: K. JINDAL[1], A. RINGLAND[1], ***L. COUTELLIER**[2];
[1]Psychology, The Ohio State Univ., Columbus, OH; [2]Psychology and Neurosci., Ohio State Univ., Columbus, OH

Abstract: The adolescent period is marked by remodeling of the prefrontal cortex (PFC), which contributes to the maturation of higher behavioral functions, including sociability. Adolescence is also a period of development during which symptoms of many psychiatric conditions arise; this includes social anxiety/withdrawal as seen in schizophrenia. The mechanisms underlying normative and pathological development of social behaviors during adolescence remain to be fully elucidated. We previously identified the transcription factor Npas4 as a major contributor to the maturational processes affecting the PFC during adolescence, suggesting that Npas4 might play a role in the acquisition of adult social behaviors. To test this idea, we first tested the sociability of Npas4 knockout (KO) mice and their wild-type (WT) controls at postnatal day (PD) 21 (pre-adolescence), PD35 (adolescence), PD70 (adulthood). Npas4 KO developed deficits in sociability during the transition from adolescence to adulthood, a phenotype not due to non-social anxiety or olfactory deficits. We then exposed C57Bl/6 mice to social isolation (SI) starting at PD22 or PD35, knowing that SI-22 but not SI-35 leads to deficits in sociability in adulthood. We showed that SI-22, but not SI-35, reduces prefrontal Npas4 mRNA levels starting mid-adolescence and into adulthood. Finally, using an AAV approach to increase Npas4 expression in the PFC, we were able to rescue SI-induced adult social deficits but only when the AAV was injected at PD22, and not at PD35. Molecular analyses reveal that the effects of SI-22 on adult social impairments might be mediated by aberrant maturation of parvalbumin interneurons due to deficits in Npas4 expression. These data indicate that prefrontal Npas4 could play an important role in the maturation of social behaviors during a critical period of prefrontal maturation, and that deficits in Npas4, as seen in the PFC of schizophrenic patients, could contribute to social withdrawal symptoms.

Disclosures: K. Jindal: None. **A. Ringland:** None. **L. Coutellier:** None.

Poster

PSTR122. Animal Models of Behavioral Dysfunction

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR122.28/B56

Topic: A.09. Adolescent Development

Support: NIH Grant AT010903

Title: Social jet lag with LED at night alters medial amygdala network and anxiety responses in adolescent mice

Authors: *P. BONILLA VILLAMIL[1], A. SHANKS[1], D. A. MCBRIDE[2], A. PORCU[1]; [1]Drug Discovery and Biomed. Sci., Univ. of South Carolina, Columbia, SC; [2]2Department of Nanoengineering, Univ. of California San Diego, San Diego, CA

Abstract: Anxiety disorders are the most common emotional disorders in adolescents. Adolescence is a remarkable period of neural plasticity in the amygdala, a key brain region regulating emotions and processing light stimuli. Adolescents commonly experience disrupted

light/dark cycles due to late sleep onset, exposure to electronic devices at night and early wake-up times on school days leading to social jet lag. Excessive LED light exposure from tables and phones has been associated with lower psychological well-being. Nevertheless, our understanding of how the adolescents' brain adapts to altered light environment remain limited. The aim of our study was to investigate the impact of social jet lag with LED light at night exposure during adolescence on medial amygdala circuitry and emotional responses. Adolescent mice were exposed for 4 weeks to either an extended light phase of 19-hrs per day with LED light appearing during the night phase for 5 days, followed by a 12-hrs light phase for the next 2 days; or control conditions consisted in 12-hrs light phase per day for 7 days. Mice were then tested for anxiety-like behaviors while simultaneously recording amygdala activation using fiber photometry. Brains were then processed for single-cell RNA-sequencing and immunofluorescence to explore the effect of altered light environment on the medial amygdala neuronal circuitry. We found that adolescent mice exposed to social jet lag with LED light at night showed increased anxiety responses and risky behaviors compared to the control group. Histological analysis revealed a significant increase in the number of somatostatin neurons and a reduction in astrocytes number. In addition, RNA-sequencing data showed changes in cell type composition and gene expression involved in synaptic communication and axonal organization in the medial amygdala. Our research provides new evidence highlighting the potential consequences of disrupted light environments during critical developmental periods in the amygdala. We suggest that social jet lag with exposure to LED light at night might represent a risk factor for developing emotional disorders, thus emphasizing the importance of maintaining consistent light environment for proper brain function and emotional regulation during adolescence.

Disclosures: P. Bonilla Villamil: None. **A. Shanks:** None. **D.A. McBride:** None. **A. Porcu:** None.

Poster

PSTR122. Animal Models of Behavioral Dysfunction

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR122.29/B57

Topic: A.09. Adolescent Development

Support: NIDA R01 DA037911
Canadian Institutes of Health Research FRN: 156272

Title: Amphetamine in adolescence induces a sex-specific mesolimbic dopamine functional phenotype in the adult prefrontal cortex

Authors: *G. HERNANDEZ[1], Z. NIU[1], J. ZHAO[1], D. MACGOWAN[1], T. CAPOLICCHIO[2], A. SONG[1], S. GUL[1], A. MOIZ[1], I. HERRERA[1], J. J. DAY[3], C. FLORES[4,1];
[1]Douglas Mental Hlth. Univ. Inst., Verdun, QC, Canada; [2]Integrated Program in Neurosci.,

montreal, QC, Canada; [3]Neurobio., Univ. of Alabama at Birmingham, Birmingham, AL; [4]Dept. of Psychiatry and Dept. of Neurol. and Neurosurg., McGill Univ., Montreal, QC, Canada

Abstract: During adolescence, the dopamine (DA) system undergoes significant structural and neurochemical changes. The Netrin-1/DCC guidance cue pathway controls protracted DA development, with environmental insults dysregulating this pathway and significantly altering prefrontal cortex (PFC) DA maturation, related cognitive processing and behavior. In male mice, recreational-like doses of amphetamine (AMPH) in early adolescence induce ectopic growth of mesolimbic DA axons to the prefrontal cortex (PFC). This effect *(i)* is mediated by drug-induced reduction in DCC receptors in mesolimbic DA axons and Netrin-1 in the nucleus accumbens (NAcc) and *(ii)* is associated with impaired inhibitory control in adulthood. Here we investigated if ectopic input of NAcc DA axons in the adult PFC leads to alteration in DA axonal varicosity expression of the dopamine transporter (DAT), in baseline DA kinetics, and in changes in DA signaling in response to the DA releaser and DAT blocker, methylphenidate (MPH). We examined if these effects could be prevented by upregulating DCC receptor expression in adolescence, using CRISPRa. Adolescent male and female mice received saline (SAL) or AMPH (4 mg/kg, i.p.) injections from PND 22 to PND 31. They were placed in distinctive compartments of a place-preference box and preference was assessed at PND 32 and again at PND 80. One day later, the DA sensor, GrabDA2h, was unilaterally microinjected into the PFC and an optical fiber was implanted. DA dynamics were measured with fiber photometry at baseline, following acute i.p. injections of SAL, and MPH (10 mg/kg). In a separate cohort of male mice, CRISPRa for *Dcc* or for *LacZ* control was microinjected into the ventral tegmental area at PND 21, followed by the AMPH or SAL treatment. Both male and female AMPH-treated mice exhibit short- and long-lasting place preference to this drug. However, in adulthood AMPH-treated males, but not females, show increased DAT density in the PFC compared to SAL controls. This male-specific increase in DAT expression is associated with *(i)* a decrease in baseline DA transients and an increase in their amplitude, *(ii)* faster DA release and a trend towards faster absorption in response to acute SAL, and *(iii)* exaggerated PFC DA signal in response to MPH. Notably, DCC upregulation prevents both the development of place preference to AMPH and alterations in adult DA dynamics. DAT expression in the PFC is significantly higher in SAL females than SAL males. The increase in DAT expression and in DA dynamics observed in the PFC of adult AMPH-treated male mice is likely to result from ectopic innervation of mesolimbic DA axons to PFC, which seem to retain the properties of their intended target.

Disclosures: G. Hernandez: None. **Z. Niu:** None. **J. Zhao:** None. **D. MacGowan:** None. **T. Capolicchio:** None. **A. Song:** None. **S. Gul:** None. **A. Moiz:** None. **I. Herrera:** None. **J.J. Day:** None. **C. Flores:** None.

Poster

PSTR122. Animal Models of Behavioral Dysfunction

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR122.30/B58

Topic: A.09. Adolescent Development

Support: NSERC CGS-M
CIHR FRN: 170130
NIDA R01 DA037911
FRN: 156272

Title: Divergentoutcomes of delta 9 - tetrahydrocannabinol in adolescence on dopamine and cognitive development in male and female mice

Authors: *T. CAPOLICCHIO[1], G. A. HERNANDEZ[2], K. ESTRADA[3], E. DUBE[3], C. FLORES[4];
[1]Intergrated Program in Neurosci., McGill, montreal, QC, Canada; [2]Douglas Mental Hlth. Univ. Institute/Mcgill, Douglas Mental Hlth. Univ. Institute/Mcgill, Verdun, QC, Canada; [3]Douglas Mental Hlth. Res. Ctr., Verdun, QC, Canada; [4]McGill Univ., Montreal, QC, Canada

Abstract: Exposure to delta 9 - tetrahydrocannabinol (THC) - the main psychoactive component of cannabis - is steadily increasing among youth. This raises concerns because core neurodevelopmental processes occur during adolescence. Dopamine axons continue to grow from the striatum towards the prefrontal cortex across adolescence, promoting the refinement of inhibitory control. This growth is controlled by the Netrin-1 guidance receptor, deleted in colorectal cancer (DCC), whose expression in dopamine neurons is regulated by the microRNA, miR-218. Our unpublished results show that repeated exposure to THC 5 mg/kg during adolescence dysregulates *Dcc* mRNA in opposite directions in male and female mice and that miR-218 is involved in *Dcc* dysregulation in males only. Here we investigated if these sex-specific molecular changes associate with differential THC effects on PFC dopamine and cognitive maturation. We treated adolescent male and female C57/Bl6 mice (postnatal day 22) with 5 intraperitoneal injections of vehicle or THC 5 mg/kg once every other day. In adulthood separate cohorts were assessed in the Go/No-go task to measure impulse control or their brains were processed to quantify the extent and organization of dopamine connectivity within the PFC. Males but not females exposed to THC show increased premature responding in the Go/No-Go task, reflective of impaired waiting impulsivity, but have fewer commission errors, indicating improved action inhibition, compared to vehicle groups. THC-treated males show a robust reduction in both the extent of the dopamine input and in the number of dopamine axon presynaptic sites in the PFC in adulthood. These changes suggest disruption to adolescent dopamine axon pathfinding. Females are protected against this effect. These data show that THC in early adolescence impacts male and female dopamine and cognitive development differently and by recruiting divergent processes. DCC-mediated events may mediate outcomes in males, but in females' compensatory mechanisms may be recruited.

Disclosures: T. Capolicchio: None. **G.A. Hernandez:** None. **K. Estrada:** None. **E. Dube:** None. **C. Flores:** None.

Poster

PSTR123. Postsynaptic Organization: Molecules, Mechanisms, and Techniques

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR123.01/B59

Topic: B.04. Synaptic Transmission

Support: Swedish VR 2022-01079
Horizon 2020 695568 SYNNOVATE
SIDB 529085
Welcome trust 202932

Title: Synaptic weight distributions are not just lognormal. Regional and age-dependent determinants.

Authors: *E. A. FRANSEN[1,2], Z. QIU[3], E. BULOVAITE[3], S. G. GRANT[4,5], M. REHN[1]; [1]Computat. Sci. and Technol., KTH Royal Inst. of Technol., Stockholm, Sweden; [2]Computat. Brain Sci., Sci. for Life Lab., Solna, Sweden; [3]Ctr. for clinical brain sciences, Univ. of Edinburgh, Edinburgh, United Kingdom; [4]Ctr. for Clin. Brain Sciences,, [5]Simons Initiative for the Developing Brain (SIDB), Ctr. for Discovery Brain Sci., Edinburgh Univ., Edinburgh, United Kingdom

Abstract: Synapses coordinate the activity of neurons, and in doing so are instrumental in behavior. They are thought to exert their influence not as individuals, but as populations. Over the course of development and learning, synaptic populations undergo structural and molecular changes, which are constrained to maintain excitation-inhibition balance, or to meet other homeostatic targets. The outcomes of this coordination can be observed in the distributions of synaptic protein content. Previous work has suggested that synaptic efficacy, as measured either by EPSPs, AMPA receptor expression, or spine volume, follows a lognormal distribution.
Here we use data from our studies of billions of synapses, where the PSD95 protein has been fluorescently labeled (Zhu et al., 2018; Cizeron et al., 2020). We investigate what statistical distributions best fit the protein data. We stratify the data based on regional boundaries and age, but also approach it in a fully data-driven way. We find that synapses from some brain regions, such as the cortex and the hippocampus, are well described by lognormal distributions, while distributions with a heavier upper tail provide a much better fit for synapses from, e.g., the midbrain and brainstem regions.
For young animals, the lognormal distribution provides a reasonable fit, consistent with previous findings (age 0-4 weeks). Regions in midbrain and brainstem however become progressively more tail-heavy during aging. This means that with aging, the synaptic populations accrue increasing numbers of very strong synapses.When aggregation of a large number of events occurs, in the presence of constraints, such as is the case in the ongoing regulation of a synaptic population, the outcome will tend to limiting distributions that maximize entropy. Importantly, it follows that when synaptic populations are indeed described by different distribution classes, the underlying processes must be distinctly different. Based on our findings, we conclude that the processes affecting synaptic parameters, including development and learning as well as homeostatic regulatory mechanisms, are sufficiently different, between brain regions, and at different ages, to lead to qualitatively distinct outcomes.

Disclosures: **E.A. Fransen:** None. **Z. Qiu:** None. **E. Bulovaite:** None. **S.G. Grant:** None. **M. Rehn:** None.

Poster

PSTR123. Postsynaptic Organization: Molecules, Mechanisms, and Techniques

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR123.02/B60

Topic: B.04. Synaptic Transmission

Support: NINDS R21NS117967
NINDS R01NS105758

Title: Electrical synapses - beyond the Connexins

Authors: J. C. MICHEL[1], ***A. MILLER**[2];
[1]Inst. of Neurosci., [2]Univ. of Oregon, Eugene, OR

Abstract: There are two main types of fast synaptic transmission in the brain - electrical and chemical. At vertebrate electrical synapses, signals pass directly through Connexin-based gap junctions between neurons, while neurotransmitter release and reception dictates chemical synapse communication. Electrical synapses are linked to a variety of neuronal disorders, are present in both the immature and mature nervous systems, are sophisticated in their communication properties, and work together with chemical synapses to articulate network function. However, there are many critical gaps in the field, one of which is that we do not have a complete 'parts list' of the electrical synapse - which we call the electrical synapse density (ESD). Understanding the proteomic makeup of the ESD would open the door to mechanistic insight into how the ubiquitous, yet understudied, electrical synapse forms and functions.
To uncover the proteomic makeup associated with neural Connexins, we used TurboID proximity labeling to 'tag' proteins near neural Connexins with biochemical 'handles', allowing us to isolate and identify the nearby proteins. We homologously recombined TurboID into the endogenous locus of a neural Connexin, and found that Connexin-TurboID localized to electrical synapses throughout the nervous system and biotinylated synaptic proteins in vivo. Using Connexin-TurboID animals, both developing larvae and adult brains, we isolated biotinylated proteins and identified them via mass spectrometry. This analysis yielded four main classes of proteins: 1) known proteins - the top two differentially biotinylated proteins were Connexin-TurboID and a cytoplasmic scaffold, ZO1; 2) adhesion regulators - ZO-related scaffolds, cell adhesion molecules, and cytoskeletal effectors; 3) trafficking proteins - Golgi, exocytic, and endocytic proteins; 4) glutamatergic synapse proteins - NMDA and AMPA receptors, postsynaptic scaffolds, and chemical synapse adhesion molecules. We hypothesize these molecules define (a) a subset of the ESD, (b) trafficking pathways that direct synaptic localization and membrane dynamics of Connexins, and (c) a striking intermingling of electrical and excitatory chemical synapse proteins. We will present our ongoing efforts using cell biological, biochemical, and functional analyses to reveal the functions of the vertebrate

electrical synapse proteome. The results shift our understanding of neural gap junctions from ‘simply Connexins’ to a complex cellular compartment that controls the structure, homeostasis, function, and plasticity of the electrical synapse and suggest an extensive interplay with chemical synapses.

Disclosures: J.C. Michel: None. **A. Miller:** None.

Poster

PSTR123. Postsynaptic Organization: Molecules, Mechanisms, and Techniques

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR123.03/B61

Topic:

Support: Intramural Research Program of the National Institute of Neurological Disorders and Stroke (NINDS) of National Institutes of Health (NIH)

Title: Modular structures in the postsynaptic density (PSD) imaged by Cryo-EM Tomography using isolated PSDs and analyzed by an automatic approach

Authors: *J. JUNG[1], X. CHEN[2], T. S. REESE[3];
[1]Natl. Inst. of Hlth., Bethesda, MD; [2]NINDS, Bethesda, MD; [3]NINDS, NIH/ NINDS, Bethesda, MD

Abstract: Postsynaptic densities (PSDs) are large protein complexes associated with the postsynaptic membrane of excitatory synapses important for synaptic function including plasticity. Conventional electron microscopy (EM) typically depicts PSDs as compact disk-like structures of hundreds of nanometers in size. Biochemically isolated PSDs were also similar in dimension revealing a predominance of proteins with the ability to polymerize into an extensive scaffold; several EM studies noted their irregular contours with often small granular structures (<30 nm) and holes. Super-resolution light microscopy studies observed clusters of PSD elements and their activity-induced lateral movement. Furthermore, our recent EM study on PSD fractions after sonication observed PSD fragments (40-90 nm in size) separate from intact PSDs; however, such structures within PSDs remained unidentified. Here we examined isolated PSDs by cryo-EM tomography with our new approach of automatic segmentation that enables delineation of substructures and their quantitative analysis. The delineated substructures broadly varied in size, falling behind 30 nm or exceeding 100 nm and showed that a considerable portion of the substructures (>38%) in isolated PSDs was in the same size range as those fragments. Furthermore, substructures spanning the entire thickness of the PSD were found, large enough to contain both membrane-associated and cytoplasmic proteins of the PSD and also similar to those by super-resolution light microscopy studies in frequency. The structures detected here appear to constitute the isolated PSD as modules of various compositions, and this modular nature may facilitate remodeling of the PSD for proper synaptic function and plasticity.

Disclosures: J. Jung: None. **X. Chen:** None. **T.S. Reese:** None.

Poster

PSTR123. Postsynaptic Organization: Molecules, Mechanisms, and Techniques

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR123.04/B62

Topic: B.04. Synaptic Transmission

Support: Marie Skłodowska-Curie Individual Fellowships 101029343

Title: Fret analysis of synaptic nanoarchitecture on a whole-brain scale in mice reveals synapse diversity of psd95 complex organisation.

Authors: *T. KAIZUKA[1], E. BULOVAITE[1], Z. QIU[2], K. MORRIS[1], M. HORROCKS[1], S. G. GRANT[1];
[2]Ctr. for Clin. Brain Sci., [1]Univ. of Edinburgh, Edinburgh, United Kingdom

Abstract: The postsynaptic terminal of excitatory synapses comprises >1,000 protein types which are assembled into multiprotein complexes. Recent studies using super-resolution microscopy reveal that postsynaptic proteins are not uniformly distributed within synapses but are packed within nanoclusters. However, super-resolution imaging of synapses is currently limited to a resolution of 20-50 nm, which limits our understanding of the packing of complexes. Here, we analyze the diversity of postsynaptic nanoarchitecture using Förster resonance energy transfer (FRET). FRET is a mechanism of energy transfer between two fluorophores that exist within close proximity (<10 nm). We evaluate FRET between PSD95, a major postsynaptic scaffold protein, labeled with two different fluorophores using HaloTag, a self-labeling protein tag that covalently binds to synthetic ligands. We labeled protein extracts, synaptosomes and histological sections of PSD95-HaloTag knock-in mouse brain with HaloTag ligands conjugated with JF552 or JFX650. Although PSD95 complexes include two copies of PSD95, FRET was not detected within these complexes in two-color coincidence detection experiments. By contrast, FRET was clearly detected in synaptosomes and synapses on histological sections, suggesting that FRET is derived from the packing of PSD95 complexes within synapses. We found synaptic heterogeneity of the FRET signal, suggesting that there is diversity between synapses in the nanometer-scale molecular distribution of postsynaptic structures. To understand the diversity of synapse nanoarchitecture across the entire brain and the lifespan, we have adapted our high-resolution, high-throughput synaptome mapping pipeline to analyze FRET data.

Disclosures: **T. Kaizuka:** None. **E. Bulovaite:** None. **Z. Qiu:** None. **K. Morris:** None. **M. Horrocks:** None. **S.G. Grant:** None.

Poster

PSTR123. Postsynaptic Organization: Molecules, Mechanisms, and Techniques

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR123.05/B63

Topic: B.04. Synaptic Transmission

Support: F31MH124283
R21MH127822
R37MH080046

Title: Direct visualization of triheteromeric NMDA receptor trafficking and organization

Authors: *M. C. ANDERSON[1], A. NIGAM[2], J. JOHNSON[2], T. A. BLANPIED[3];
[1]Univ. of Maryland Sch. of Med. Program In Neurosci., Baltimore, MD; [2]Univ. of Pittsburgh, Pittsburgh, PA; [3]Dept. of Physiol., Univ. of Maryland Sch. of Med., Baltimore, MD

Abstract: N-methyl-d-aspartate receptors (NMDARs) constitute one of the most common ionotropic glutamate receptors in the brain and play crucial roles in synaptic plasticity, brain development, and cell health. NMDARs are heterotetramers comprised of two obligatory GluN1 subunits and two GluN2A-D or GluN3A-B subunits, allowing for a multiplicity of possible receptor subtypes. In the hippocampus, the most common NMDAR subtypes are GluN2A or GluN2B diheteromeric (containing two GluN2A or two GluN2B subunits) or GluN2A/GluN2B triheteromeric (containing one GluN2A and one GluN2B subunit) NMDARs. Importantly, the subunit composition of NMDARs plays important roles in cellular and synaptic organization, trafficking, channel kinetics, and signaling capabilities. Indeed, compared to their diheteromers, triheteromeric NMDARs display intermediate agonist sensitivity, open channel probability and deactivation kinetics. Additionally, triheteromeric NMDARs contain multiple C-tails which provide the unique ability to participate in a variety of downstream signaling pathways. Although triheteromeric NMDARs make up potentially even a majority of NMDARs in the brain, there are currently no ways to visually distinguish triheteromeric NMDAR from their diheteromeric counterparts. Thus, their subcellular distribution remains mysterious and the mechanisms for receptor trafficking are unknown, representing a substantial gap in our current understanding of NMDAR biology. Here, we developed a tool utilizing biomolecular complementation to specifically visualize triheteromeric NMDARs. This tool utilizes split reporter proteins fused to the N-terminal domains of GluN2A and GluN2B which allows for complementation and fluorescence detection only when GluN2A and GluN2B complex together in a tetrameric NMDAR. Importantly, these split-tagged NMDARs display no aberrations in agonist and antagonist sensitivity and traffic avidly to hippocampal synapses. To expand this toolbox and address questions surrounding triheteromeric NMDAR trafficking we took advantage of a split-HaloTag molecule which becomes catalytically active when complemented in a triheteromeric NMDAR. We combined HaloTag ligands with multiple spectral properties and membrane permeability to elucidate factors that control triheteromeric NMDAR trafficking, subcellular organization, and synaptic content. These results suggest a broad approach to measure and functionally control specific subtypes of heteromeric receptors, including and beyond NMDARs.

Disclosures: M.C. Anderson: None. **A. Nigam:** None. **J. Johnson:** None. **T.A. Blanpied:** None.

Poster

PSTR123. Postsynaptic Organization: Molecules, Mechanisms, and Techniques

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR123.06/C1

Topic: B.04. Synaptic Transmission

Support: T32 Neuroscience Predoctoral Training Grant NS099042
NIH Grant R35NS116879
NINDS Grant UF1NS107710
NIH Grant R01MH119154
NIH Grant F31NS130979

Title: Subsynaptic ampa and gaba-a receptor nanodomains assemble in the absence of neurotransmitter release

Authors: *H. J. RAMSAY[1], S. E. GOOKIN[1], A. M. RAMSEY[1], D. J. KAREEMO[1], K. C. CROSBY[1], D. G. STICH[2], S. S. OLAH[1], H. S. ACTOR-ENGEL[1], K. R. SMITH[1], M. J. KENNEDY[1];
[1]Dept. of Pharmacol., [2]Advanced Light Microscopy Core (ALMC), Univ. of Colorado, Anschutz Med. Campus, Aurora, CO

Abstract: At both excitatory and inhibitory synapses, neurotransmitter receptors and their associated protein scaffolds congregate into distinct, nanometer-scale subsynaptic domains (SSDs). Interestingly, postsynaptic receptor SSDs are located immediately adjacent to presynaptic active zones. These transsynaptic molecular assemblies are thought to increase the efficiency of neurotransmission by concentrating postsynaptic receptors directly across the synaptic cleft from sites of presynaptic neurotransmitter release. Previous studies have characterized how activity affects the number, size and transsynaptic alignment of postsynaptic receptor SSDs at mature synapses. However, it remains unknown whether the release of neurotransmitter is required for the initial assembly of receptor SSDs prior to or during synapse development. Here, we chronically blocked neurotransmitter release before synaptogenesis using lentivirally expressed tetanus neurotoxin. We then evaluated synaptic nano-architecture using multiple forms of super resolution microscopy. In agreement with previous work, excitatory and inhibitory synapses formed in the absence of neurotransmitter release. While silencing reduced the overall size of excitatory and inhibitory postsynaptic specializations, synaptic transmission was not required at any point for AMPARs, GABA(A)Rs or their respective scaffold proteins to assemble into SSDs. Furthermore, AMPAR SSDs maintained alignment with the presynaptic active zone protein RIM1. Thus, assembly of receptors and their respective scaffolds into transsynaptically aligned nanostructures, is an intrinsic synaptic property which can be further refined by subsequent activity-dependent mechanisms. Experimental model / rigor statement: Cultured neurons from both male and female P0-P1 Sprague-Dawley rat pups were pooled and used for all experiments. Biological sex differences were not assessed. In this exploratory work, we ensured scientific rigor through the use of appropriate controls, biological and technical replicates with sufficient sample size, and blinding when possible.

Disclosures: **H.J. Ramsay:** None. **S.E. Gookin:** None. **A.M. Ramsey:** None. **D.J. Kareemo:** None. **K.C. Crosby:** None. **D.G. Stich:** None. **S.S. Olah:** None. **H.S. Actor-Engel:** None. **K.R. Smith:** None. **M.J. Kennedy:** None.

Poster

PSTR123. Postsynaptic Organization: Molecules, Mechanisms, and Techniques

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR123.07/C2

Topic: B.04. Synaptic Transmission

Support: DA R33041876
Department of Pharmacology and Toxicology
Stark Neurosciences Research Institute
Strategic Research Initiative

Title: Spinophilin-dependent regulation of protein phosphatase 1 targeting and postsynaptic density protein organization in the striatum and hippocampus

Authors: E. T. CLAEBOE[1], A. BEIRAGHI SALEK[3], *A. BAUCUM II[2];
[1]Pharmacol. and Toxicology, [2]Indiana Univ. Sch. of Med., Indianapolis, IN; [3]Biol., Indiana University-Purdue Univ. Indianapolis, Indianapolis, IN

Abstract: Neuroplasticity, or the ability of neuronal cells to respond to a changing environment, requires alterations in synaptic function within the brain. Biochemical changes, such as the dynamic regulation of protein function, can be mediated by protein phosphorylation; specifically, balancing the actions of serine/threonine protein kinases and their highly promiscuous serine/threonine counterpart, protein phosphatases. Protein phosphatase-1 (PP1) and similar proteins account for more than 90% of serine/threonine phosphatase activity in eukaryotes. Of note, PP1 complexes with over 100 targeting subunits and may form over 500 holoenzyme complexes comprised of targeting, regulatory, and substrate proteins along with the PP1 catalytic subunit (PP1c). PP1 holoenzyme complexes provide substrate specificity by modulating the subcellular targeting or/and activity of the catalytic subunits. The neurabins (spinophilin and its homologue neurabin) are the two major postsynaptic density (PSD)-enriched PP1 targeting proteins. We and others have found that the neurabins are critical for striatal-dependent motor outputs, including motor learning, locomotor sensitization to psychostimulants, and repetitive motor outputs associated with neurodiverse presentations such as obsessive-compulsive spectrum disorders. Therefore, these proteins are critical mediators of striatal function. However, how specifically they target PP1 to the PSD and PSD-associated substrates is not known. To begin to assess the role of the neurabins in targeting PP1 and its substrates to striatal PSDs, we have validated a PSD isolation protocol and found enrichment of the canonical PSD marker, PSD-95, with a lack of immunoblotting for the cytosolic protein, GAPDH. Using a crude fractionation protocol, we recently found that loss of spinophilin decreased total synaptic protein expression in crude synaptic fractions in the hippocampus, including a decrease in expression of

the GluN2B subunit of the NMDA receptor. Differences in protein expression of PP1 alpha and gamma as wells as PSD proteins, such as ionotropic glutamatergic receptors within hippocampus and striatum of WT mice and global spinophilin KO mice will be discussed.

Disclosures: E.T. Claeboe: None. **A. Beiraghi Salek:** None. **A. Baucum II:** None.

Poster

PSTR123. Postsynaptic Organization: Molecules, Mechanisms, and Techniques

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR123.08/C3

Topic: B.04. Synaptic Transmission

Support: Pew Charitable Trust
FONDECYT 11230291
NINDS

Title: Voltage imaging of synaptic activity from neocortical pyramidal neurons in vivo

Authors: *V. CORNEJO, R. YUSTE;
Columbia Univ., New York, NY

Abstract: Excitatory synapses located in dendritic spines play a critical role in dendritic integration, making their function essential for brain function. However, the precise mechanisms of dendritic computation and input-output transformation *in vivo* remain poorly understood due to the lack of direct electrical measurement in dendritic spines. Recent evidence suggests that spine-dendrite coupling has the potential to electrically modify synaptic potentials. In this study, our aim was to investigate further the membrane potential dynamics of spines and dendrites in pyramidal neurons from somatosensory cortex of mice *in vivo* using two-photon imaging and genetically encoded voltage indicators (GEVIs). We imaged a GEVI specifically targeted to spines and dendrites, called postASAP. By employing high frame rate scanning and improving the signal-to-noise ratio through time-locked activity, we achieved high spatiotemporal precision in capturing the dynamic behavior of spines and dendrites of somatosensory cortex. To activate synaptic events, we employed presynaptic stimulation techniques using bipolar electrodes or optogenetic stimulation with ChRmine. This enabled us to isolate and observe small voltage changes occurring within spines and their relationship to parent dendrites. Our observations demonstrated fast and precise activation of individual spines upon presynaptic stimulation, providing evidence for the independent nature of spine activation. Importantly, we found that spine voltages remained compartmentalized after optogenetic presynaptic activation of individual spines, highlighting their persistent role as elementary voltage compartments. These findings advance our understanding of the regulation of dendritic signaling and electrical function in spines. The precise temporal characterization of subthreshold events provides valuable insights into the dynamics of dendritic integration and synaptic processing.

Furthermore, these findings could have important implications for unraveling the underlying mechanisms of synaptic plasticity and synaptic dysfunctions.

Disclosures: V. Cornejo: None. **R. Yuste:** None.

Poster

PSTR123. Postsynaptic Organization: Molecules, Mechanisms, and Techniques

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR123.09/C4

Topic: B.04. Synaptic Transmission

Title: Shn-1 coordinates nociceptive synaptic transmission and aversive behavior in Caenorhabditis elegans

Authors: A. R. GENTILIN, *J. A. ZAHRATKA;
Neurosci., Baldwin Wallace Univ., Berea, OH

Abstract: SHANK is a family of scaffold proteins responsible for the organization of glutamate receptors at postsynaptic densities. Specifically, *SHANK3* mutation results in aberrant development of glutamatergic synapses and is implicated in neurodevelopmental disorders including autism spectrum disorders and Phelan-McDermid Syndrome. Interestingly, patients with *SHANK3* mutations exhibit multiple phenotypes including hypotonia, sleep issues, and increased pain tolerance, likely due to mislocalization of glutamate receptors and other postsynaptic proteins. In the present study, we explore the structural and functional effects of SHANK mutation in a nociceptive circuit using the nematode *Caenorhabditis elegans*. SHN-1, the *C. elegans* homolog of SHANK3, shares sequence similarity and functional domains with vertebrate counterparts, suggesting conserved function. We found that two different *shn-1* mutations decrease responses to noxious stimuli sensed by the ASH sensory neurons. ASHs are glutamatergic nociceptors capable of sensing chemical, mechanical, and osmotic stimuli that synapse onto downstream interneurons resulting in backward locomotion away from the stimulus (“reversal”), a type of avoidance behavior. In the octanol avoidance assay for chemical stimuli, *shn-1* mutants fail to demonstrate serotonin- or food-mediated stimulation of aversive responses, suggesting that SHN-1 activity is required for proper modulation of responses to chemorepellents. Intriguingly, *shn-1* mutants have reduced responses to light nose touch independently of serotonin, but food stimulates the frequency of reversals, implying differential modulation of this sensory modality. Localization studies of AMPA receptors in the downstream AIB and AVA interneurons are ongoing, but we hypothesize that we will see mislocalization of these receptors in both *shn-1* mutant alleles. Together, these results suggest conservation of glutamatergic synapse organization and allow for greater understanding of SHANK-mediated disruption of sensation in neurodevelopmental disorders.

Disclosures: A.R. Gentilin: None. **J.A. Zahratka:** None.

Poster

PSTR123. Postsynaptic Organization: Molecules, Mechanisms, and Techniques

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR123.10/C5

Topic: B.04. Synaptic Transmission

Support: Cure Sanfilippo Fellowship Grant
MPS Society Grant

Title: Dysfunction of mitochondria and synaptic organization contribute to Sanfilippo syndrome pathology

Authors: *J. ACEVEDO, P. J. MATHEWS, L. E. POLGREEN, M. IACOVINO;
The Lundquist Inst. for Biomed. Innovation at Harbor-UCLA Med. Ctr., Torrance, CA

Abstract: Mucopolysaccharidosis type III (MPS III), or Sanfilippo syndrome, is a fatal lysosomal storage disease caused by deficiency in enzymes specifically involved in the degradation of heparan sulfate (HS) and other and other glycosaminoglycans (GAGs). There are four variants of MPS III, distinguished by the specific enzyme deficiency. MPS III is characterized by severe neurological symptoms (e.g., cognitive and learning deficits) linked to neurodegeneration and premature death in patients. While HS and GAGs in the lysosomes has been proposed to underlie the neuropathology, recent evidence from animal models points to mechanisms occurring early in development, especially those involved in mitochondrial and synaptic function as playing a major role. Thus, to mechanistically define the early pathology in MPS III, we performed proteomic analyses to identify which proteins are abnormally expressed in MPS III patient-derived iPSC brain organoids and differentiated forebrain neurons compared to healthy controls (HC). We found an array of proteins dysregulated in the MPS III-derived cells and validated a subset with western blots and immunofluorescence. We found several proteins involved in mitochondrial function dysregulated, including a 2-fold increase in the translocase of the mitochondrial outer membrane 40 (TOMM40). This dysfunction was further corroborated morphologically, as MPS III forebrain neurons in culture appear abnormal, displaying a punctuated or aggregated form, suggesting a defect in mitochondrial function. Identified were also several dysregulated proteins involved in synaptic function, including Neuronal pentraxin 2 (PTXN2), Neurexin 2 (NRXN 2), Ephrin-B2 (EFNB2) and Teneurin transmembrane protein 2 (TENM2), all of which showed a significant increase (about 2-fold) compared to controls. At the post-synaptic level, Neuroligin-2, Glypican-1, Glypican-4, and glutamate receptor 1 (GluR1) and GluR3, were also upregulated 4- to 8-fold in MPS III cells compared to HC. We investigated AMPA receptor expression via immunofluorescence and found it significantly upregulated ($p < 0.05$) in MPS III forebrain neurons compared to WT. Overall, our data suggest dysfunction in the mitochondria, and synaptic formation and organization of MPS III neurons compared to controls.

Disclosures: J. Acevedo: None. **P.J. Mathews:** None. **L.E. Polgreen:** None. **M. Iacovino:** None.

Poster

PSTR123. Postsynaptic Organization: Molecules, Mechanisms, and Techniques

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR123.11/C6

Topic: B.04. Synaptic Transmission

Title: The role of PPP2R5D and its human de novo mutations in liprin-α1 liquid-liquid phase separation and synaptic transmission

Authors: *A. MAYER[1], A. GANGULY[1], R. TARTAVARDY[1], V. JANSSENS[2], R. HONKANEN[3], S. STRACK[4], H. XIA[1];
[1]Univ. of Rochester, Rochester, NY; [2]KU Leuven, Leuven, Belgium; [3]Univ. of South Alabama, Mobile, AL; [4]Univ. of Iowa, Iowa City, IA

Abstract: Human *de novo* mutations in the gene encoding PPP2R5D (R5D), a regulatory subunit of protein phosphatase 2A (PP2A), results in Jordan's Syndrome (PPP2R5D-related intellectual disability). PP2A is a serine/threonine phosphatase responsible for about half of dephosphorylation events in mammalian cells. The PP2A holoenzyme is composed of a scaffolding "A" subunit, a regulatory "B" subunit, and a catalytic "C" subunit. R5D is highly expressed in the brain; yet, how R5D regulates PP2A localization and catalytic activity, and how *de novo* mutations alter holoenzyme function to regulate neuronal processes is unknown. Knock-in mouse models with a R5D mutation (E198K or E420K) have increased action potential (AP) firing. In addition, E420K mice have increased mEPSC frequency, leading us to explore R5D's role in synaptic transmission. Liprin-α1, an excitatory synaptic scaffolding protein, interacts with R5D, thus suggesting R5D functions at excitatory synapses. We aim to explore how R5D-PP2A regulates liprin-α1 via dephosphorylation and how *de novo* mutations in R5D alter the function of liprin-α1 at synapses. R5D interacting proteins, including liprin-α1, contain at least one short linear motif (SLiM): LxxIxE. Liprin-α1 has two SLiMs (SLiM 1 and SLiM 4) which we mutated separately or together and transfected into WT HEK 293 cells. We hypothesized liprin-α1 mutants (SLiM 1m, SLiM 4m, or SLiM 1m/SLiM 4m) would have increased phosphorylation when transfected into HEK 293 cells. Liprin-α1 SLiM 4m and Liprin-α1 SLiM 1m/SLiM 4m transfected HEK 293 cells resulted in an increased percentage of cells with the formation of droplet-like structures. We measured fluorescence recovery after photobleaching on liprin-α1 SLiM 4m droplets and found a significant fraction of droplets are mobile, with a recovery half time of approximately 10 seconds. Our data suggest phosphorylation of liprin-α1 triggers the formation of these droplets. Other PPP2R5 family B subunits recognize this SLiM on liprin-α1, so we utilized R5D knock-out (KO) HEK 293 cells to assess the contribution of R5D to liprin-α1 droplet formation. R5D KO cells transfected with liprin-α1 showed a significant increase in cells with droplets, suggesting R5D is the primary B subunit targeting PP2A to liprin-α1 ($n=3$, $p<0.0001$, ANOVA). We are actively determining the phosphorylation sites on liprin-α1 responsible for its droplet formation. In parallel, we are testing our prediction that human PPP2R5D *de novo* mutations affect synaptic transmission by regulating liprin-α1

phosphorylation. In summary, our work will help determine the molecular mechanism for R5D and its human mutations in excitatory synaptic transmission.

Disclosures: **A. Mayer:** None. **A. Ganguly:** None. **R. Tartavardy:** None. **V. Janssens:** None. **R. Honkanen:** None. **S. Strack:** None. **H. Xia:** None.

Poster

PSTR123. Postsynaptic Organization: Molecules, Mechanisms, and Techniques

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR123.12/C7

Topic: B.04. Synaptic Transmission

Support: 1ZIANS003140-09

Title: Functional analysis of an ASD-associated of NLGN4X within the dimerization domain

Authors: *E. HONG, K. ROCHE;
NIH/NINDS, Bethesda, MD

Abstract: Autism spectrum disorder (ASD) is a multifaceted neurodevelopmental disorder characterized by challenges in social interactions, deficits in communication, and repetitive behaviors. There are many genes that have been identified as being associated with ASD from both human genetics and etiological evidence. Neuroligins (NLGN1-3, NLGN4X, and NLGN4Y) are highly associated with ASD. NLGNs are postsynaptic cell adhesion molecules that interact with presynaptic neurexins (NRXNs) and play a crucial role in neuronal development, synaptic transmission, and synaptic plasticity. Among the NLGN family members, NLGN4X stands out as having the highest number of variants associated with ASD. Most of the rare variants associated with ASD are found within the extracellular esterase homology domain and those mutations display deficits in surface trafficking and impaired neuronal function. Here, we report a novel NLGN4X variant, T439I, identified in ASD patients and located in the dimerization domain. NLGNs can form both homodimers and heterodimers, and dimerization may play a critical role in their function in neural networks, although research on this aspect is limited. Using biochemical approaches, we have characterized NLGN4X T439I, which displays impaired oligomerization with NLGN3 without affecting its stability, glycosylation, or homo-dimerization. Additionally, we discovered a reduced interaction with NRXN1beta(+S4), which is known to play a role at inhibitory synapses. Based on these results, we propose a model in which NLGN4X T439I impacts the balance between excitatory and inhibitory neurotransmission.

Disclosures: **E. Hong:** None. **K. Roche:** None.

Poster

PSTR123. Postsynaptic Organization: Molecules, Mechanisms, and Techniques

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR123.13/C8

Topic: B.04. Synaptic Transmission

Support: 1ZIANS003140-08

Title: Neuroligin-3 ectodomain cleavage results in multiple species of intracellular domain products

Authors: *K. MCDANIEL[1,1], J. JEONG[1,2], Y. LI[1,3], K. W. ROCHE[1];
[1]Natl. Inst. for Neurolog. Disorders and Stroke, NIH, Bethesda, MD; [2]Amyloid Solution Inc, Seongnam, Korea, Republic of; [3]Natl. Inst. for Neurolog. Disorders and Stroke, Proteomics Core Facility, Bethesda, MD

Abstract: Neuroligins (NLGNs) are a family of post-synaptic adhesion molecules that, along with pre-synaptic neurexins, aid in the formation, function, and maintenance of synapses. There are five NLGN family members (1-4X/4Y) with different functions and localization. NLGN3 has greater than 98% homology between mice and humans and is the only of the five NLGNs that is localized to both excitatory and inhibitory synapses. Additionally, many mutations in NLGN3 have been identified in people with autism spectrum disorders (ASDs) and these mutations have had a variety of phenotypes in cellular studies. Despite its characterization as a canonical adhesion molecule, our group and others have shown that NLGN3 can be cleaved by metalloproteases in the ectodomain (ECD) region proximal to the transmembrane domain. While the NLGN3 ECD has been studied as a potent mitogen in pediatric glioblastomas, the physiological role of NLGN3 post-cleavage has yet to be researched. We are investigating the physiological role of the NLGN3 ECD, as well as the NLGN3 intracellular domain (ICD) that remains post-cleavage. Previously, our group showed that there are multiple cleavage pathways that differ with cellular activity levels as modeled by treatment with PMA, a PKC pathway activator. We have discovered multiple ICD species differentiated by treatment with protease inhibitors and that these species are differentially enriched for phosphorylation at S725, a Cdk5 phosphorylation site. Using mass spectrometry, we have identified a putative basal cleavage site upstream of the transmembrane domain of NLGN3 that could be necessary for the creation of multiple NLGN3 ICD species. These results establish the physiological role of NLGN3 cleavage and provide the basis for work on pathogenic mutations to improve therapeutic treatments for ASD.

Disclosures: K. McDaniel: None. **J. Jeong:** None. **Y. Li:** None. **K.W. Roche:** None.

Poster

PSTR123. Postsynaptic Organization: Molecules, Mechanisms, and Techniques

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR123.14/C9

Topic: B.04. Synaptic Transmission

Support: HFSP LT000737/2019-C
EMBO ALTF 860-2018
Joachim Herz Stiftung 850027

Title: Synaptic machinery for protein homeostasis

Authors: *C. SUN;
Mol. Biol. and Genet., Univ. of Aarhus, Aarhus, Denmark

Abstract: An individual neuron hosts over 10000 synapses in a complex dendritic and axonal arbor. To edit the local proteomes of individual synapses, both protein synthesis and degradation machinery are decentralized to synapses. Using single-molecule localization microscopy, we quantified the copy number and assembly state of both the protein synthesis machine (ribosomes) and the major protein degradation machine (proteasomes) in neuronal dendrites and synapses. We mapped the machinery for protein synthesis, their newly-synthesized protein product, as well as their synaptic distribution during both synaptic maintenance and plasticity and found a significant correlation between synaptic calcium activity and new protein supply. We further reveal a surprising abundance of free proteasome regulatory (19S) particles near synapses. These free 19S proteasome particles regulate synaptic transmission independently of the proteasome. This suggests an emerging idea that complex protein machines adapt to subcellular needs and take on 'moonlighting' function at brain synapses.

Disclosures: C. Sun: None.

Poster

PSTR123. Postsynaptic Organization: Molecules, Mechanisms, and Techniques

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR123.15/C10

Topic: B.04. Synaptic Transmission

Support: NIH R00 MH118425
NARSAD BBRF Young Investigator Award

Title: Understanding the localization and function of endogenous MDGA1 at excitatory synapses in the hippocampus

Authors: *M. A. SANDOVAL, L. ACOSTA SOTO, J. DIAZ-ALONSO;
Anat. & Neurobio., UC Irvine, Irvine, CA

Abstract: MAM domain-containing glycosylphosphatidylinositol anchor 1 (MDGA1) is a completely extracellular, GPI-anchored synaptic adhesion molecule. In humans, the MDGA1 gene is associated with schizophrenia. Mouse MDGA1 knockouts (KO) lack gross anatomical defects, but show impaired excitation/inhibition balance and defects in cognitive function.

Overexpression and crystal structure studies have presented MDGA1 as one of the few known synaptic repressors acting via a direct, negative modulation of another synaptic adhesion molecule, NLGN2, at inhibitory synapses. However, proximity-based proteomic data suggest that endogenous MDGA1 localizes to excitatory, not inhibitory synapses. Given these apparently contradictory findings, a reliable analysis of MDGA1 localization is essential to gain understanding about its physiological role; however, protein-level analyses have been hampered by the lack of a suitable antibody. Here, we generated a knock-in mouse line expressing HA-tagged MDGA1. Utilizing this model, we quantified the colocalization of immunolabeled HA-MDGA1 puncta with four pre- and postsynaptic markers (presynaptic: vGluT1 and vGAT; postsynaptic: Homer1b/c and NLGN2) in area CA1 and Dentate Gyrus (DG) of the hippocampus (n = 5/8 mice/marker). Knock-in and wildtype samples of both sexes were processed at the peak of MDGA1 expression on postnatal day 15 (P15). In CA1, we found HA-MDGA1 colocalized significantly more with excitatory postsynaptic protein Homer1b/c compared to inhibitory postsynaptic protein (and putative functional binding partner) NLGN2. In both CA1 and DG, HA-MDGA1 colocalized more with excitatory presynaptic marker vGluT1 compared with vGAT, albeit non-significantly. Together, these data indicate that, in vivo, MDGA1 localizes predominantly to excitatory postsynapses, not inhibitory synapses as previously described. Ongoing work is focused on assessing MDGA1's underlying role at excitatory synapses via sparse, CRISPR-mediated KO of MDGA1. We are analyzing the effects of KO on excitatory synaptic transmission and LTP at CA3->CA1 synapses through dual whole-cell patch clamp recordings in acute mouse slices. Overall, our data expands our understanding of the localization and function of endogenous MDGA1, an enigmatic protein relevant to neuropsychiatric disorders.

Disclosures: M.A. Sandoval: None. **L. Acosta Soto:** None. **J. Diaz-Alonso:** None.

Poster

PSTR124. Homeostatic Plasticity

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR124.01/C11

Topic: B.05. Synaptic Plasticity

Support: NIH Grant R01-NS-065992

Title: Altered homeostatic plasticity in the spinal cord of Fragile X mouse

Authors: *D. PEKALA[1], P. A. WENNER[2];
[1]Emory Univ., Atlanta, GA; [2]Cell Biol., Emory Univ. Sch. of Med., Atlanta, GA

Abstract: Fragile X Syndrome (FXS) is the most common form of inherited intellectual disability. It is caused by a loss-of-function of the FMR1 gene on the X chromosome, resulting in the absence of fragile X messenger ribonucleoprotein (FMRP). Altered cortical activity is an underlying pathology of FXS that is associated with sensory hypersensitivity. Normally,

networks maintain activity levels within an appropriate range through a set of homeostatic plasticity mechanisms including compensatory adjustments in synaptic strength and/or intrinsic excitability. Previous work has suggested that multiple forms of homeostatic plasticity are impaired in FXS models. FXS and other forms of autism spectrum disorder (ASD) are also associated with significant delays in motor development, yet spinal cord studies in these model systems are surprisingly rare. Here we assess one form of homeostatic plasticity in spinal neurons in the FXS mouse model (Fmr1 KO). We have recently shown that blocking NMDA receptor-mediated miniature postsynaptic currents (mPSCs) in spinal neurons of the chick embryo triggers a fast compensatory increase in the amplitude of AMPAergic mPSCs. This rapid form of AMPAergic scaling has also been identified in hippocampal cultures and slices illustrating the fundamental importance of this kind of plasticity. Therefore, in the present study we have extended this work to the spinal network of the genetically advantageous mouse model system. We asked whether this form of homeostatic plasticity was altered in the neonatal Fmr1 KO mouse. We found that FXS spinal neurons express an exaggerated form of rapid AMPAergic scaling when compared to WT littermates. These results suggest that altered homeostatic plasticity in the developing spinal cord could contribute to impaired motor development in this ASD model.

Disclosures: **D. Pekala:** None. **P.A. Wenner:** None.

Poster

PSTR124. Homeostatic Plasticity

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR124.02/C12

Topic: B.05. Synaptic Plasticity

Title: Robust automated detection of spontaneous synaptic events using deep learning

Authors: *I. DELVENDAHL[1], P. O'NEILL[1], M. BACCINO-CALACE[1], P. RUPPRECHT[2], M. MUELLER[1];
[1]Dept. of Mol. Life Sci., Univ. of Zurich, Zurich, Switzerland; [2]Brain Res. Inst., Zurich, Switzerland

Abstract: Synapses are crucial components of neural circuits, serving as both structural and functional units that determine neuronal connectivity and communication. Quantitative information about synaptic transmission is therefore key to our understanding of neural function. Spontaneous subthreshold synaptic events carry important information about synaptic function and neuronal computation. However, due to their stochastic nature and low signal-to-noise ratio, reliable and consistent localization of these events in neurophysiological data remains highly challenging. Here, we present a novel method for detecting spontaneous synaptic events utilizing a deep convolutional neural network termed miniML. miniML uses supervised learning with training data from a large number of miniature excitatory postsynaptic current recordings from a cerebellar synaptic preparation. We train a deep learning model that achieves high-performance

classification with accuracy exceeding 98%. Application of the trained classifier to electrophysiological time-series data accurately detects synaptic events under a broad range of signal-to-noise conditions. Using simulated ground-truth data, we demonstrate that miniML outperforms commonly used detection methods regarding both precision and recall. Deep-learning-based detection is not threshold dependent and robust to false positives. We use synaptic recordings from different species and preparations to test the generalizability of miniML. Our results show that the analysis method can be easily applied to diverse datasets, either using a pre-trained model or via transfer learning if event kinetics differ strongly from the original training data. Across different synaptic preparations, miniML thus enables automated, reproducible, and precise analysis of subthreshold synaptic events. Running in Python and with the possibility to easily train new models, the analysis method is also highly flexible. As a robust and generalizable analysis tool, miniML will facilitate the analysis of subthreshold synaptic events and open new avenues to study the synaptic basis of neural function.

Disclosures: I. Delvendahl: None. **P. O'Neill:** None. **M. Baccino-Calace:** None. **P. Rupprecht:** None. **M. Mueller:** None.

Poster

PSTR124. Homeostatic Plasticity

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR124.03/C13

Topic: B.05. Synaptic Plasticity

Title: A Dynamin-Dependent Increase in Quantal Size Maintains Synaptic Transmission after Tetanic Stimulation

Authors: A. NAIR[1], ***N. BOLLMOHR**[2,1,3], P. MUTTATHUKUNNEL[1,2,3], M. MÜLLER[1,2,3,4]; [1]Univ. of Zurich, Zurich, Switzerland; [2]Dept. of Mol. Life Sci., Zurich, Switzerland; [3]Neurosci. Ctr. Zurich, Zurich, Switzerland; [4]Univ. Res. Priority Program "Adaptive Brain Circuits in Develop. and Learning" (AdaBD), Zurich, Switzerland

Abstract: Some synapses reliably transmit information during intense synaptic activity. Yet, the mechanisms supporting robust synaptic transmission during ongoing activity remain enigmatic. Here we show that tetanic stimulation of the *Drosophila* neuromuscular junction induces a prominent decrease in quantal content for several minutes after stimulation. The magnitude and exponential time course of this post-tetanic quantal content decrease correlates with stimulation frequency and duration. Tetanic stimulation reduces the number of release-ready synaptic vesicles assessed by electrophysiological analysis. Accordingly, electron microscopy revealed a lower number and density of synaptic vesicles at stimulated synapses. Tetanic stimulation also decreases the abundance of the presynaptic scaffold Bruchpilot. By contrast, release probability and presynaptic calcium influx are unchanged. Intriguingly, despite the reduction in synaptic vesicle number, evoked synaptic responses are largely unaltered after the tetanus. The post-tetanic decrease in synaptic vesicle number is paralleled by an increase in quantal size. This post-

tetanic quantal size increase is attenuated after application of the non-competitive dynamin inhibitor dynasore, or presynaptic expression of the temperature-sensitive *dynamin* mutant allele *shibire(ts)*. Moreover, evoked amplitudes are reduced following tetanic stimulation upon dynasore treatment or presynaptic *shibire(ts)* expression. By contrast, the post-tetanic quantal size increase persists after genetic GluRIIA or GluRIIB subunit ablation, receptor calcium permeability impairment or postsynaptic calcium channel block. Together, our findings suggest that a dynamin-dependent increase in quantal size maintains synaptic transmission after tetanic stimulation.

Disclosures: A. Nair: None. **N. Bollmohr:** None. **P. Muttathukunnel:** None. **M. Müller:** None.

Poster

PSTR124. Homeostatic Plasticity

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR124.04/C14

Topic: B.05. Synaptic Plasticity

Support: European Research Council Grant 724866
Israel Science Foundation Grant 1663/18
Volkswagen Foundation and the Ministry of Science and Culture of Lower Saxony grant ZN3458
The Deutsche Forschungsgemeinschaft (440813539 and 448865644)
BIRAX Regenerative Medicine Initiative (46BX18TKIS)
the Israel Ministry of Science, Technology and Space, and Rosetrees Trust (A2590)

Title: Nmda receptors maintain homeostatic firing rate set points in hippocampal circuits

Authors: *A. RUGGIERO[1], L. R. HEIM[1], I. SHAPIRA[1], M. KATSENELSON[1], D. HREAKY[1], K. ROSENBLUM[2], I. SLUTSKY[1];
[1]Tel Aviv Univ., Ramat Aviv, Israel; [2]Sagol Dept Neuro, Univ. of Haifa, Haifa, Israel

Abstract: What molecular mechanisms do neurons use to homeostatically maintain their activity at a given set point? While NMDA receptors (NMDARs) are the classic, central regulators of Hebbian-like synaptic plasticity, their role in homeostatic plasticity has remained controversial. Utilizing long-term multi-electrode array recordings in hippocampal networks *ex vivo*, we found that sustained inhibition of NMDARs by structurally different blockers, including ketamine, rapidly and stably suppressed mean firing rate (MFR), while maintaining homeostatic responses to activity perturbations. Interestingly, ketamine reduced MFR set point through an intrinsic, but not synaptic, mechanism that ultimately lowers the excitation/inhibition (E/I) ratio. This mechanism requires eEF2K and BDNF. Importantly, chronic local delivery of NMDAR blockers to the CA1 suppressed MFR set point across vigilance states in behaving mice. Altogether, our

results indicate that the degree of NMDAR activation dictates homeostatic MFR set point. These results highlight NMDARs as a network-wide MFR set-point modulator and extend NMDAR function beyond its canonical role in synaptic plasticity. Moreover, this study raises the possibility that some NMDAR-dependent behavioral effects are mediated by modified homeostatic set points.

Disclosures: A. Ruggiero: None. **L.R. Heim:** None. **I. Shapira:** None. **M. Katsenelson:** None. **D. Hreaky:** None. **K. Rosenblum:** None. **I. Slutsky:** None.

Poster

PSTR124. Homeostatic Plasticity

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR124.05/C15

Topic: B.05. Synaptic Plasticity

Support: NIH Grant R01MH070727
NIH Grant R01MH066198
NIH Grant T32MH064913

Title: Probing mechanisms of homeostatic synaptic plasticity using chemogenetics

Authors: *E. BAGATELAS[1], E. KAVALALI[2];
[1]Neurosci., [2]Pharmacol., Vanderbilt Univ., Nashville, TN

Abstract: Synaptic connections in the central nervous system are stable, but also flexible and dynamic to activity changes throughout one's lifetime. Neuronal homeostasis is a tightly regulated mechanism that enables neurons to self-regulate their activity in response to sustained changes in network activity as well as synaptic plasticity. Homeostatic synaptic plasticity is critical in this context for inducing adjustments within a dynamic range to maintain balanced function. It is induced by prolonged perturbations that produce signaling cascades that elicit uniform scaling of synaptic inputs across a neuron. In this study, we aimed to trigger homeostatic synaptic scaling using chemogenetic tools—Designer Receptors Exclusively Activity by Designer Drugs (DREADDs) —in a molecularly specific manner to modulate neuronal activity. We chose these chemogenetic-based tools since they are engineered to be activated upon binding to synthetic ligands, such as Compound 21. We used hM4D(Gi) DREADDs to selectively suppress activity in excitatory neurons in our primary cell culture to recapitulate a low-activity environment. Based on our initial observations, we are currently focusing to parse out what post-synaptic mechanisms are key players in driving homeostatic synaptic plasticity in this setting.

Disclosures: E. Bagatelas: None. **E. Kavalali:** None.

Poster

PSTR124. Homeostatic Plasticity

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR124.06/C16

Topic: B.05. Synaptic Plasticity

Support: R01065992

Title: The role of Na^+-K^+ATPase activity in the rapid homeostatic regulation of spontaneous network activity in the chick embryo spinal cord.

Authors: C. E. GONZALEZ-ISLAS[1,2], ***P. WENNER**[1];
[1]Emory Univ. Sch. of Med., Atlanta, GA; [2]Doctorado en Ciencias Biologicas, Univ. Autonoma de Tlaxcala, Tlaxcala, Mexico

Abstract: The chick embryo spinal cord exhibits spontaneous bursts of network activity (SNA) that drive limb movements, similar to the fetal movements in humans. This activity is a result of the highly excitable nature of the developing synaptic circuit, where GABAergic neurotransmission is depolarizing and excitatory. Previous studies have demonstrated that blocking either excitatory GABAergic or glutamatergic transmission in the living chick embryo temporarily inhibits the movements generated by SNA in the spinal cord. However, these embryonic movements begin to recover within two hours and are completely restored after 12 hours of continuous receptor blockade. The mechanisms responsible for this early recovery have remained unclear. Recently, we have identified a homeostatic mechanism that involves a robust compensatory response occurring shortly after blocking either GABAergic or glutamatergic neurotransmitter receptors. In the isolated cord *in vitro*, we observed a significant depolarization of the resting membrane potential (RMP) in motoneurons and interneurons within the first two hours of receptor blockade. These changes in RMP result in a reduction of the threshold current required for neuronal spiking, which persists in the presence of the antagonist. Accordingly, we proposed that a rapid depolarization in RMP serves as a crucial mechanism for restoring network activity following neurotransmitter receptor blockade. Previous work has demonstrated that a hyperpolarizing conductance driven by activation of the Na^+-K^+ ATPase could influence RMP and is activated by significant spiking activity, as occurs during SNA. In this study, we explore the role of the Na^+-K^+ ATPase in facilitating rapid changes in RMP to homeostatically regulate SNA. Further, we have begun to test whether intracellular Na^+ and/or Ca^{++} dynamics associated with SNA could regulate the Na^+-K^+ ATPase activity and therefore play a role in the homeostatic regulation of SNA.

Disclosures: C.E. Gonzalez-Islas: None. **P. Wenner:** None.

Poster

PSTR124. Homeostatic Plasticity

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR124.07/C17

Topic: B.05. Synaptic Plasticity

Support: SFARI

Title: Homeostatic plasticity and learning in the primary visual cortex (V1) of SCN2A$^{+/-}$ rats

Authors: *N. DIAZ RODRIGUEZ[1], E. BOUGIOUKLI[2], G. TURRIGIANO[3];
[1]Brandeis Univ. Grad. Neurosci. Program, Waltham, MA; [3]Brandeis Univ., [2]Brandeis Univ., Waltham, MA

Abstract: During development, neocortical circuits must balance the need to change in response to experience (learning) with the need to maintain stable function. Homeostatic plasticity serves to stabilize activity in neocortical circuits in the face of learning-induced perturbations caused by Hebbian forms of plasticity, to enable both flexibility and stability. Our laboratory has shown that genes associated with autism spectrum disorders (ASDs), including *Shank3*, exhibit homeostatic plasticity deficits in the primary visual cortex (V1), and impairments in a vision-dependent learned behavior in which rats learn to hunt crickets (prey capture). Because several ASD-associated genes are known to have impairments in homeostatic plasticity, we hypothesized that homeostatic plasticity within V1, and learning of this vision-dependent behavior, might be impaired in other rat models of ASD. Here, we test this hypothesis in a rat model of haploinsufficiency for *SCN2A*, which encodes for a voltage-gated sodium channel (Nav1.2). To mimic human haploinsufficiency, we used Long-Evans rats that were heterozygous for *SCN2A* knockout (*SCN2A*$^{+/-}$), and experiments were performed during the classic visual system critical period, when visual cortical circuitry is especially plastic. To induced homeostatic plasticity, we chronically suppressed neuronal activity for 24 hours using inhibitory DREADDS (Designer Receptor Exclusively Activated by Designer Drugs) expression in binocular V1 (V1b), followed by *ex vivo* acute slice electrophysiology recordings from L2/3 pyramidal neurons of wildtype (WT) and *SCN2A*$^{+/-}$ rats to measure intrinsic excitability. For the prey capture paradigm, animals underwent three acclimation days with immobilized crickets, followed by three days of cricket hunting, and then a retention test seven days later. Our electrophysiology data demonstrate that L2/3 pyramidal neurons in V1b of *SCN2A*$^{+/-}$ rats have a higher intrinsic excitability than WT littermates, but are unable to engage intrinsic homeostatic plasticity to increase excitability upon DREADDs inhibition. Surprisingly, preliminary data suggest that during the prey capture paradigm *SCN2A*$^{+/-}$ rats show no obvious impairment in prey capture learning or retention; further analysis will reveal whether there are differences in strategy between genotypes. These results show that SCN2A is required for intrinsic homeostatic plasticity in L2/3 pyramidal neurons but does not impair vision-dependent learning, raising the possibility that other forms of homeostatic plasticity are able to compensate for the loss of intrinsic homeostatic plasticity.

Disclosures: **N. Diaz Rodriguez:** None. **E. Bougioukli:** None. **G. Turrigiano:** None.

Poster

PSTR124. Homeostatic Plasticity

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR124.08/C18

Topic: B.05. Synaptic Plasticity

Support: Canadian Institutes of Health Research

Title: Endogenous TNF mitigates morphine induced locomotor sensitization and synaptic plasticity in the nucleus accumbens

Authors: *D. ARNOUIL[1], M. COTT[2], S. VALADE[2], D. STELLWAGEN[3];
[1]McGill University, Montreal, Montreal, QC, Canada; [3]Ctr. Res. Neurosci, [2]McGill Univ., Montreal, QC, Canada

Abstract: The opioid crisis has demonstrated the need for new therapeutic strategies, but despite increasing research in the field, the plasticity mechanism induced by opioid addiction in the reward circuitry are far from being understood. In the Nucleus Accumbens (NAc), a key structure of the reward system, it is now clear that glial cells are activated by drugs of abuse such as cocaine or morphine and contribute to the development of addictive behavior.
We have previously shown that repeated administration of cocaine activates striatal microglia and induces the production of the inflammatory cytokines Tumor Necrosis factor alpha (TNF), which in turn depresses glutamatergic synaptic strength and mitigate the cocaine induced locomotor sensitization. Whether this phenomenon is specific to cocaine or can be generalized to other drugs was not known. As opioids also activate microglia, we investigated whether TNF could similarly regulate the effects of opioid sensitization. At the behavioural level, we observed similar TNF regulation as we previously observed with cocaine. Blocking TNF signaling results in an increase in locomotor sensitization. Further, re-activating microglia via a TLR4 agonist after a period of abstinence can suppress sensitization. Thus, TNF appears to suppress both the development and expression of sensitization. However, the plasticity at glutamatergic synapses in the NAc following morphine treatment are more complex and not as well characterized as those following cocaine. Here we present data on the synaptic changes both in NAc core and shell following repeated morphine treatment.
Our finding show that despite morphine treatment causes the same behavioural sensitization as cocaine, the synaptic changes underlying this behaviour are likely different. This gives us new insight on the plasticity mechanisms involved in opioid addiction and underline the role of TNF in regulating it. Importantly the fact that TLR4 activation is capable of mitigating morphine induce locomotor sensitization place this receptor as a potential therapeutic target in the treatment of opioid addiction.

Disclosures: D. Arnouil: None. **M. Cott:** None. **S. Valade:** None. **D. Stellwagen:** None.

Poster

PSTR124. Homeostatic Plasticity

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR124.09/C19

Topic: B.05. Synaptic Plasticity

Support: NIH Grant 1R01MH130600

Title: Biogenesis of calcium-permeable AMPA receptors via GluA2 mRNA editing in homeostatic synaptic plasticity

Authors: *L. PETERSON[1], H. MAN[2];
[1]Pharmacol., [2]Biol., Boston Univ., Boston, MA

Abstract: During neuronal activity deprivation, synaptic strength is upregulated by homeostatic synaptic plasticity (HSP) via an increase in synaptic accumulation of AMPA receptors (AMPARs). Calcium permeable AMPARs (Cp-AMPARs) have been shown to be required for the initiation of HSP, however, how Cp-AMPARs are generated in the early stage of HSP remains less clear. Calcium permeability of AMPARs is determined by the presence of GluA2, which is normally edited at the mRNA level by ADAR2. This editing event converts a glutamine to an arginine in the ion pore region of the GluA2 protein, rendering GluA2(R)-containing receptors calcium impermeable. We found that in primary cortical neurons, activity silencing by tetrodotoxin (TTX) led to a suppression in GluA2 editing and an increase in the expression of Cp-AMPARs. Influx of AMPAR-gated calcium was blocked by overexpression of the editing enzyme ADAR2, suggesting that increased GluA2(Q) mediates this change in AMPAR calcium permeability. Consistently, overexpression of ADAR2 blocked the TTX-induced homeostatic increase in synaptic GluA1. Since ADAR2 has multiple substrates, we used a CRISPR-Cas13 system to selectively increase editing efficacy of GluA2, which also blocked the expression of HSP. To examine the mechanisms underlying activity-dependent GluA2 editing, we found that ADAR2 nuclear expression was decreased by TTX incubation. In an *in vivo* mouse model of HSP in visual deprivation, similar changes were observed in the visual cortex including increased GluA2(Q) expression and decreased ADAR2 nuclear expression. Also, brain viral expression of ADAR2 was sufficient to abolish the change in GluA2 editing and block HSP expression in the visual cortex after binocular deprivation. These findings indicate that GluA2 editing is a major molecular event in the formation of Cp-AMPARs for the expression of HSP.

Disclosures: L. Peterson: None. **H. Man:** None.

Poster

PSTR124. Homeostatic Plasticity

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR124.10/C20

Topic: B.05. Synaptic Plasticity

Support: NIMH MH70727
NIMH MH064913
2T32MH064913-17

Title: The role of MeCP2 in the molecular action of ketamine and its implication for the treatment of Rett syndrome

Authors: *M. K. PIAZZA[1], E. T. KAVALALI[2], J. L. NEUL[3], L. M. MONTEGGIA[4];
[2]Vanderbilt Univ., [1]Vanderbilt Univ., Nashville, TN; [3]Neurosciences, Univ. of California San Diego, La Jolla, CA; [4]Vanderbilt Brain Inst., Vanderbilt Brain Inst., Nashville, TN

Abstract: Ketamine has received attention in recent years for its rapid and sustained action as an antidepressant, even in patients with treatment resistant depression. Systemic ketamine administration in mice blocks NMDA receptors (NMDAR) at rest to ultimately cause an increase in BDNF protein production and an associated increase in excitatory synaptic strength in the hippocampus, underlying its antidepressant action. The long-term effects of ketamine necessitate phosphorylation of the transcription regulator Methyl-CpG binding protein 2 (MeCP2), but the role of MeCP2 in the acute effects of ketamine require further study. Interestingly, mutations in the *MECP2* gene underlie the neurodevelopmental disorder Rett syndrome, and BDNF expression has been shown to be impaired in both human patients and mice modeling Rett syndrome. Therefore, it is plausible that there may be a convergence between these two disparate disorders and ketamine may hold therapeutic potential in Rett syndrome. We sought to further investigate the function of MeCP2 in the molecular action of ketamine and its implications for neurodevelopmental and psychiatric disease using a combination of electrophysiological, behavioral and biochemical techniques in mutant mouse lines. The findings of this study may shed light on ketamine's modulation of excitatory/inhibitory balance in the hippocampus in both a healthy condition and in the context of Rett syndrome.

Disclosures: M.K. Piazza: None. **E.T. Kavalali:** None. **J.L. Neul:** None. **L.M. Monteggia:** None.

Poster

PSTR124. Homeostatic Plasticity

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR124.11/C21

Topic: B.05. Synaptic Plasticity

Title: Functional description of metaplasticity at excitatory synapses

Authors: *G. BUSETTO[1], B. G. MOCKETT[2], W. C. ABRAHAM[2];
[1]Dept. of Neurosci., Univ. of Verona, Verona, Italy; [2]Dept. of Psychology, Univ. of Otago, Dunedin, New Zealand

Abstract: Long-term potentiation (LTP) of excitatory inputs is considered a neural mechanism of learning. LTP is a positive feed-back process and may saturate its effects, resulting in the occlusion of further modulation of synaptic function and, consequently, of learning. Hence there is a need for the homeostatic adjustment of the threshold for LTP induction. One described mechanism is "metaplasticity": for example, a priming period of high frequency postsynaptic action potential activity (pAP-prime) before LTP induction reduces LTP expression, as predicted by the Bienenstock Cooper Munro theory of 1982. However, later experiments designed at testing the effect of pAP-prime have given conflicting results, with LTP being inhibited, facilitated or both. The dependance itself of metaplasticity on the presence of action potentials has been questioned. Moreover, a detailed description of the functional conditions in which metaplasticity may extert its homeostatic control over LTP is missing. We are aiming to shed light on metaplasticity functionality by testing different pAP-prime frequencies (30, 60 and 90Hz,) and different time delays between pAP-prime and LTP induction (0.5, 2 and 10min) in whole-cell patch clamp recordings of EPSPs evoked in CA1 hippocampal neurons of acute rat brain slices after Schaffer collateral electrical stimulation. On average, LTP expression is suppressed only when induced 2 or 10min after a 60Hz pAP-prime (EPSP amplitude measured 30min after LTP induction, relative to baseline: 2min delay, 1.1±0.13, n=8; 10min delay, 1.1±0.14, n=9; LTP only: 1.7±0.08, n=8; p = 0.003 and 0.004, respectively), but not in the other conditions tested so far (60Hz, 0.5min delay: 1.4±021, n=6, p=0.3; 90Hz, 2 and 10min delays: 1.4±0.23, n=9, p=0.3; 1.5±0.21, n=3, p=0.4; respectively). However, in all primed conditions a relevant percentage of tested neurons show no LTP (range 33-87%, cutoff at 1.3 relative to baseline), a data to be confronted with LTP-only experiments (0%). Conclusions: i) postsynaptic firing activity does exert a homeostatic control of future LTP; ii) this effect appears to be modulated by the frequency of firing and by the time delay with the LTP induction; iii) CA1 hippocampal neurons might be a non-homogeneous group of neurons as far as it concerns their sensitivity to metaplasticity.

Disclosures: G. Busetto: None. **B.G. Mockett:** None. **W.C. Abraham:** None.

Poster

PSTR124. Homeostatic Plasticity

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR124.12/C22

Topic: B.05. Synaptic Plasticity

Support: NSF Grant 1900212

Title: Modulation of Synaptic Neurotransmitter Levels by Auto-receptor in Caenorhabditis elegans.

Authors: *R. FORMISANO;
Delaware State Univ., Dover, DE

Abstract: Modulation of Synaptic Neurotransmitter Levels by Auto-receptor in *Caenorhabditis elegans.*

Authors*R. FORMISANO; Delaware State University, Dover, DE, United States.

DisclosuresR.Formisano: None.

Abstract

Precise modulation of neurotransmitters is critical for neural function. Mechanisms of synaptic vesicular fusion and neurotransmitter clearance are highly controlled processes whose finely-tuned regulation is essential for the functioning of the nervous system. For example, levels of the catecholamine neurotransmitter dopamine released into the synaptic cleft critically influence a wide range of neural functions, ranging from motor control to cognition, learning, motivation and reward. The fundamental cellular and molecular processes that influence synaptic neurotransmitter levels are known to be conserved across phylogeny. The complexity of mammalian CNS circuits makes it particularly challenging to disentangle the inter-digitated mechanisms controlling dopamine release. The well-defined nervous system of *Caenorhabditis elegans* provides a simple yet powerful model that has been successfully used to link genes, individual neurons and neural circuits to specific behaviors. The overall goal of our research is to understand *how neuronal function and communication are regulated by the molecular interactions that maintain levels of synaptic dopamine release and reuptake.* We have utilized *C.elegans* as our model organism to better understand the cellular roles of two modulators of synaptic feedback for the neurotransmitter dopamine (DA): the pre-synaptic DA auto-receptor DOP-2 and the DA membrane transporter DAT-1. Based on functional imaging of individual synaptic termini, our work has shown that the D2-like auto-receptor DOP-2 provides a feedback loop in modulating synaptic vesicle fusion rates. In addition, we found that the DA transporter DAT-1 provides another feedback loop for synaptic DA modulation. Based on these results, we seek to explore how the DAT-1 and DOP-2 proteins interact in order to allow us to fill the gaps in our understanding of how DA release and reuptake are regulated in *C. elegans.* At this stage, we are specifically interested in a transient receptor potential (TRP) channels which are involved in vision, taste, mechano-sensation, hearing, osmosensation, thermosensation and phototransduction with downstream effects that include learning and memory. We present, preliminary results of our studies to test the interaction between the *TRP channel, trp-4 and the DOP-2 auto-receptor in the modulation of the levels and activity of dopamine at synapses in C.elegans.*

Disclosures: R. Formisano: None.

Poster

PSTR124. Homeostatic Plasticity

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR124.13/C23

Topic: B.05. Synaptic Plasticity

Support: NIH Grant AG062222

Title: Preferential Upregulation of the ProSAAS Chaperone During Hippocampal Homeostatic Scaling

Authors: S. MITIAS, S. NAIR, N. SCHAFFER, *I. LINDBERG;
Neurobio., Univ. of Maryland Sch. of Med., Baltimore, MD

Abstract: To maintain network stability in the face of elevated activity, neurons engage a negative feedback mechanism termed homeostatic scaling in order to preserve relative synaptic weight. In proteomics studies by others, the small secretory chaperone proSAAS was found to be one of the most highly regulated proteins in a cell culture model of homeostatic signaling. Our own prior work has demonstrated that proSAAS exhibits anti-aggregant behavior against alpha synuclein and Abeta fibrillation *in vitro* and is upregulated in cell models of proteostatic stress. Nigral proSAAS overexpression provides profound dopaminergic cell protection in an animal model of Parkinson's disease. The specific role that this protein might play in homeostatic scaling, however, is not yet clear. To learn more about proSAAS regulation, we compared its expression in a neuronal model of homeostatic scaling (primary hippocampal cell culture) to other synaptic components using Western blotting and qPCR. Upon treatment with the sodium channel blocker tetrodotoxin, proSAAS levels increased by 144% ± 15% (mean ± SD; n=5 independent experiments) relative to the vehicle control, while we saw only a 115% ± 12% increase in carboxypeptidase E (CPE) levels, and no change in 7B2 (also known as Scg5) levels (95% ± 11%). Upon treatment with the GABA antagonist bicuculline, proSAAS protein levels decreased to 38% ± 8.6% of the vehicle control group, while CPE levels were 99% ± 18%, and 7B2 levels were 96% ± 7.1% of the vehicle control. There was no significant change in HSP90 or HSP70 levels upon drug treatment. mRNA changes were not as robust as protein changes. Immunocytochemistry and immunohistochemistry were used to visualize the location of proSAAS immunoreactivity both in cell culture and in coronal mouse sections. Confocal analysis supports the synaptic localization of a portion of neuronal proSAAS and demonstrates that tetrodotoxin-induced changes also occur within the cell body, most likely in the Golgi region. Finally, immunohistochemical analysis of the mouse hippocampus shows that immunoreactive proSAAS is especially concentrated within the mossy fiber layer, in a pattern similar to Timm staining. We hypothesize that dynamic changes in proSAAS expression may play an important role in synaptic proteostatic processes.

Disclosures: **S. Mitias:** None. **S. Nair:** None. **N. Schaffer:** None. **I. Lindberg:** None.

Poster

PSTR124. Homeostatic Plasticity

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR124.14/C24

Topic: B.05. Synaptic Plasticity

Support: European Research Council 724866
Israel Science Foundation 1663/18

Deutsche Forschungsgemeinschaft 440813539 and 448865644
BIRAX Regenerative Medicine Initiative 46BX18TKIS

Title: Homeostatic regulation of CA1 firing rate set points and contextual memory retrieval in mice

Authors: *R. ATSMON[1], K. BAR-OR[1], D. MANDIL[1], I. SLUTSKY[2];
[1]Physiol. and Pharmacology, Sackler Fac. of Med., Tel Aviv Univ., Tel aviv, Israel; [2]Tel Aviv Univ., Tel Aviv Univ., Tel Aviv, Israel

Abstract: Homeostatic mechanisms stabilize activity of neural circuits by keeping firing rates in a given circuitry within a set-point range. Accumulated evidence suggests that mean firing rate (MFR) represents a physiological variable regulated by homeostatic systems in cultured neuronal networks ex vivo and in primary visual cortex in vivo. However, whether the hippocampus, known to be highly engaged in memory encoding and storage, is capable of homeostatic regulation of MFR in response to a chronic perturbation is unknown. Furthermore, whether homeostatic regulation of MFR impacts hippocampus-dependent behavior remains elusive. To address these questions, we applied long-term single-unit recordings in freely behaving mice to measure spontaneous spiking activity of CA1 pyramidal neurons under the baseline spontaneous activity and in response to chronic chemogenetic perturbation (Gq-DREADD) of hippocampal interneurons. Clozapine N-oxide (CNO) was applied via osmotic pump for five days to achieve a constant perturbation. Our results show that MFRs are regulated by vigilance states, displaying a down-regulation by low-arousal states. Activation of Gq-signaling in interneurons led to an acute suppression of MFR across a population of CA1 pyramidal cells. Despite the constant perturbation, the activity of CA1 excitatory neurons gradually recovers after 3 days, returning to the original MFR set-point value. Interestingly, MFR returned to its brain state-specific set-point value. At the sub-population level, low-firing rate CA1 neurons lost their positive modulation by NREM sleep, while high-firing rate neurons preserved negative modulation by NREM sleep during perturbation. While active wakefulness, NREM and REM sleep all contributed to renormalization of MFR to a set point, REM was the only state that actively promoted upward MFR homeostasis. Mechanisms that restore MFR at the population level also restore contextual memory retrieval in fear conditioned mice. This study provides the first direct evidence on homeostatic regulation of MFR set points by brain states and points to the role of homeostatic mechanism in preserving a core function of the hippocampus to retrieve stored memories.

Disclosures: **R. Atsmon:** None. **K. Bar-Or:** None. **D. Mandil:** None. **I. Slutsky:** None.

Poster

PSTR124. Homeostatic Plasticity

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR124.15/C25

Topic: B.05. Synaptic Plasticity

Support: NIH Grant R01EY025613

Title: Learning-induced modulation of Primary Binocular Visual Cortex (V1b) activity in freely behaving rats during a cricket hunting prey-capture paradigm

Authors: *D. P. LEMAN[1], B. J. LANE[2], B. A. CARY[4], G. TURRIGIANO[3];
[2]Biol., [3]Dept of Biol., [1]Brandeis Univ., Waltham, MA; [4]Biol., Brandeis Univ. Grad. Neurosci. Program, Waltham, MA

Abstract: Neurons in the primary visual cortex (V1) have mean firing rates that span several orders of magnitude, and individual neurons regulate their activity around stable mean firing rates that they return to following chronic visual perturbations. These findings suggest that V1 neuron have an individual 'firing rate set point' that is maintained through homeostatic plasticity mechanisms. We wondered whether these set points are truly 'set' or might be malleable when animals participate in a more salient ethological experience. To investigate this, we used a prey-capture paradigm in which juvenile rats learn to chase, capture, and consume live crickets. Rodents are opportunistic omnivores with an innate drive to hunt but must learn to hunt effectively; we observed a progressive improvement in their performance across multiple hunting sessions in a single day, indicating robust learning. To determine whether this behavior depends on visual cortex we expressed excitatory Designer Receptors Exclusively Activated by Designer Drugs (DREADDs) in parvalbumin-positive interneurons in V1 to suppress V1 activity acutely during hunting and found that this dramatically decreased performance. Next, we used chronic, in vivo extracellular recordings in V1b to track neuronal activity during learning. Initially a small subset of neurons in V1b were significantly modulated during pursuit epochs, but there was a dramatic increase in the number of pursuit-responsive neurons as the animal's performance improved. Next, we measured how learning impacted mean neuronal firing rates on longer timescales after hunting to assess whether firing rate set points were altered by learning. Our preliminary analysis indicates that V1b neurons have stable baseline firing rates prior to hunting, but that these mean rates change slowly over many hours post-learning. These changes persist in both individual neurons and at the population level for at least 36 hours following learning, suggesting that this salient experience causes long-term changes in neuronal and network excitability. Taken together, our preliminary data show that V1 plays a crucial role in prey capture learning, that learning induces dramatic changes in the responsivity of V1b neurons, and that learning can perturb mean and population level activity for days after the last hunting session. Our future work will provide insight into how Hebbian and homeostatic plasticity mechanisms interact to generate these dramatic learning-induced changes in V1b.

Disclosures: D.P. Leman: None. **B.J. Lane:** None. **B.A. Cary:** None. **G. Turrigiano:** None.

Poster

PSTR124. Homeostatic Plasticity

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR124.16/C26

Topic: B.05. Synaptic Plasticity

Support: NIH Grant NS130108
NIH Grant NS085164

Title: How discrete homeostatic signals stabilize synapse function across time

Authors: *C. FRANK[1], B. MALLIK[1], K. M. LEMBKE[2], D. J. ZWIEFELHOFER[1];
[1]Univ. of Iowa, Iowa City, IA; [2]George Fox Univ., Newburg, OR

Abstract: Neuroplasticity is needed to respond to exogenous stress. It implies change on structural or functional levels. Yet synaptic systems must also retain stability and keep within appropriate physiological bounds. Forms of homeostatic plasticity are well-documented in synaptic systems. Years ago, it was posited that homeostatic processes at synapses needed to be slow and long-lasting, to buffer against deleterious challenges to function. But recent work demonstrates that homeostatic signals work both on rapid, short-term timescales and chronic, long-term maintenance timescales. We know discrete homeostatic signals, but we have little understanding of how the identified molecules integrate into a coherent control system across phases of time. The *Drosophila melanogaster* neuromuscular junction (NMJ) is an ideal synapse to untangle this problem. The NMJ has robust homeostatic capacity. One way is through a process termed "presynaptic homeostatic potentiation" (PHP). With PHP, NMJ neurotransmitter receptors are genetically or pharmacologically impaired. This type of challenge causes decreased NMJ quantal size, but it also triggers retrograde, muscle-to-nerve signaling that drives increased transmitter release (quantal content). As a result, normal levels of muscle excitation are maintained. Challenged with impaired receptor function, an NMJ can induce and execute PHP in just ten minutes. But PHP can also be maintained for long periods of developmental time.Our group has developed genetic and pharmacological tools to understand how discrete homeostatic signals stabilize NMJ function across time. We have organized and studied PHP in three different phases. **1) Induction.** We can slow down the short-term induction of PHP using loss-of-function conditions in chaperone-encoding genes *hsp26* and *hsp27*. With slowed induction, we have characterized sequences of signals needed for the induction of PHP. One finding is that we can slow down the expression of PHP and correlate the timing to enhancements of discrete active zone proteins. **2) Transition.** We engineered a way to monitor a transition from PHP induction to maintenance using the glutamate receptor inhibitor 1-naphthyl acetyl spermine (NASP). NASP can be fed directly to flies or applied directly to the NMJ to induce homeostatic signaling. We have tested how induction signals yield to previously identified maintenance factors, like Heartless (Htl)/Fibroblast Growth Factor Receptor, Src64B, and Target of Rapamycin (TOR). **3) Maintenance.** We uncoupled the induction and maintenance phases of PHP. One finding is that an IP_3-directed calcium store release pathway sustains stable NMJ outputs over time.

Disclosures: C. Frank: None. **B. Mallik:** None. **K.M. Lembke:** None. **D.J. Zwiefelhofer:** None.

Poster

PSTR124. Homeostatic Plasticity

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR124.17/C27

Topic: B.05. Synaptic Plasticity

Support: DFG Grant CRC/TRR 167

Title: Impact of anticonvulsant medication on excitatory synaptic strength - unveiling homeostatic plasticity in the adult human cortex

Authors: *A. EICHLER[1,2], M. LENZ[1,2], H. LU[2], P. KRUSE[2], J. STRÄHLE[3,4], S. DIAZ[6], P. TURCO[7], H. HEMELING[2], P. STÖHR[2], I. VIDA[7], J. BECK[3,4,5], A. VLACHOS[2,5,8];
[1]Inst. of Neuroanatomy and Cell Biol., Hannover Med. Sch., Hannover, Germany; [2]Dept. of Neuroanatomy, Inst. of Anat. and Cell Biology, Univ. of Freiburg, Freiburg, Germany; [3]Dept. of Neurosurg., Med. Ctr. and Fac. of Medicine, Univ. of Freiburg, Freiburg, Germany; [4]Ctr. for Advanced Surgical Tissue Analysis (CAST), [5]Ctr. for Basics in Neuromodulation (NeuroModulBasics), Fac. of Medicine, Univ. of Freiburg, Freiburg, Germany; [6]Simulation Lab. Neuroscience, Jülich Supercomputing Center, Inst. for Advanced Simulation, Jülich, Forschungszentrum Jülich, Jülich, Germany; [7]Inst. of Integrative Neuroanatomy and NeuroCure Cluster of Excellence, Charité-Universitätsmedizin Berlin, Berlin, Germany; [8]Ctr. Brain Links Brain Tools, Univ. of Freiburg, Freiburg, Germany

Abstract: Homeostasis, a fundamental principle in biological systems, is crucial for maintaining functional physiological processes. In the human body, numerous regulatory mechanisms have been described to uphold the homeostasis of the system. However, the precise mechanisms and relevance of homeostatic synaptic plasticity in the human brain remain largely unknown, despite being a well-known principle observed in animal models. In this study, we aimed to investigate the effects of antiepileptic drugs on neurotransmission and functional properties of layer 2/3 pyramidal cells in the adult human cortex. Using whole-cell patch-clamp recordings, transcriptome analysis, confocal imaging and modeling of neuronal networks, we demonstrated that antiepileptic drugs, such as lamotrigine, which attenuate network activity, subsequently induce compensatory strengthening of excitatory neurotransmission in both murine and human cortex. These changes are accompanied by transcriptomic and structural changes. Our findings provide the first evidence of homeostatic plasticity in the adult human cortex induced by the clinical application of antiepileptic drugs. Furthermore, they offer a potential mechanistic explanation for the recurrence of seizures following withdrawal from antiepileptic drug treatment.

Disclosures: **A. Eichler:** None. **M. Lenz:** None. **H. Lu:** None. **P. Kruse:** None. **J. Strähle:** None. **S. Diaz:** None. **P. Turco:** None. **H. Hemeling:** None. **P. Stöhr:** None. **I. Vida:** None. **J. Beck:** None. **A. Vlachos:** None.

Poster

PSTR124. Homeostatic Plasticity

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR124.18/C28

Topic: B.05. Synaptic Plasticity

Support: HHMI Gilliam Fellowship
R35NS111562

Title: Shank3 Phosphorylation Regulates Synaptic Scaling Up Via Homer1 and mGluR5

Authors: *A. A. GUERRERO, C.-H. WU, V. TATAVARTY, G. G. TURRIGIANO; Brandeis Univ., Waltham, MA

Abstract: Shank3 is a postsynaptic scaffold protein important in properly localizing receptors and other synaptic proteins at the excitatory synapse to allow for its proper functioning. Mutations in Shank3 are strongly associated with Autism Spectrum Disorders (ASD). Shank3 is necessary for the expression of synaptic scaling, a form of homeostatic plasticity that bidirectionally modulates post-synaptic strength in response to activity perturbations, stabilizing neuronal activity. The molecular mechanisms by which Shank3 regulates synaptic scaling are not yet known. In cultured cortical neurons, our lab recently identified two sites on Shank3 (S1586 and S1615) that show a persistent hypo-phosphorylation during activity blockade and transient hyper-phosphorylation in response to an increase in activity. Using electrophysiology and immunocytochemistry, we demonstrated that introducing Shank3 phosphorylation (phosphomimetic mutant) blocks scaling up while preventing Shank3 phosphorylation (phosphodeficient mutant) abolishes scaling down. Our data thus suggest that the phosphorylation state of these two sites seem to act as a bidirectional switch permissive to scale in a particular direction in response to changes in activity. Here we aim to further investigate the molecular machinery downstream of Shank3 dephosphorylation that allows for synaptic scaling up. To this end, we found that enhancing metabotropic glutamate receptor 5 (mGluR5) activity by using a positive allosteric modulator rescued scaling up in neurons expressing the Shank3 phosphomimetic mutant. Notably, mGluR5 has been shown to associate with Shank3 through Homer1, a postsynaptic protein also crucial for scaling and known binding partner of Shank3. Further, we found that the phosphomimetic mutant of Shank3 has reduced interaction with Homer1, suggesting that phosphorylation of Shank3 disrupts Shank3-Homer1-mGluR5 interactions to prevent scaling up. Future experiments aim to elucidate the downstream mechanism by which mGluR5 activation rescues scaling up.

Disclosures: A.A. Guerrero: None. **C. Wu:** None. **V. Tatavarty:** None. **G.G. Turrigiano:** None.

Poster

PSTR124. Homeostatic Plasticity

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR124.19/C29

Topic: B.05. Synaptic Plasticity

Support: NIH Grant NS117372
NIH Grant NS121284
SFARI Award 551354
Brain and Behavior Research Foundation Young investigator Award 27792

Title: Postsynaptic Matrix Metalloproteinase 2 Controls Trans-Synaptic Homeostatic Plasticity in Drosophila

Authors: *Y. CAI[1], T. WANG[1], J. CHEN[2], A. CHAKRABORTY[3], T. CUI[1], T. WANG[1,4]; [1]Pharmacol. & Physiol., [2]Sch. of Hlth., [3]Dept. of Biol., [4]Interdisciplinary Program in Neurosci., Georgetown Univ., Washington, DC

Abstract: Homeostatic plasticity is a form of synaptic regulation that stabilizes neural function. At the *Drosophila* neuromuscular junction (NMJ), when postsynaptic glutamate receptor function is inhibited pharmacologically or genetically, an increase of presynaptic neurotransmitter release compensates for the postsynaptic perturbation so that the excitation is maintained at a stable level in the postsynaptic cell. This form of homeostatic regulation is termed Presynaptic Homeostatic Potentiation (PHP). PHP can be induced acutely by application of a glutamate receptor antagonist philanthotoxin-433 or chronically by genetic deletion of glutamate receptor subunit GluRIIA. PHP is highly conserved across species, ranging from *Drosophila* to humans. In our previous studies, we found that a glial-secreted extracellular matrix (ECM) protein, Multiplexin, is critical for both acute induction and chronic maintenance of PHP. *Drosophila* Multiplexin, homologue to mammalian collagen XV/XVIII, is a soluble collagen in the ECM. Endostatin, the C-terminal domain of Multiplexin, can be proteolytically cleaved and released from Multiplexin. Intriguingly, Endostatin serves as an intercellular communication signal to facilitate neurotransmitter release during PHP through presynaptic calcium channels. However, the identity of the protease that cleaves Multiplexin to release the signaling molecule Endostatin remains to be elucidated. In this study, we investigated the function of matrix metalloproteases 1 and 2 (*Mmp1* and *Mmp2*), the only two *Mmps* in *Drosophila*, in the rapid induction and long-term maintenance of PHP. Using CRISPR genome editing, we generated a molecular null mutant allele of *Mmp2*. We systematically examined the function of *Mmp1* and *Mmp2* in PHP using electrophysiological, immunohistochemistry, biochemistry, confocal-imaging, and super-resolution imaging methods. Our data suggest that *Mmp2*, but not *Mmp1* is necessary for both acute induction and chronic maintenance of PHP. In addition to our genetic evidence, we further validated our finding by pharmacological inhibitors of Mmps. Using tissue-specific RNAi knockdown and rescue approaches, we also demonstrated that Mmp2 functions specifically in the postsynaptic muscle to control the trans-synaptic homeostatic plasticity. Moreover, we showed that the Endostatin domain of Multiplexin is proteolytically cleaved and released by Mmp2 for the rapid induction of PHP. In summary, we have uncovered the identity and function of an ECM protease, Mmp2, which increases presynaptic neurotransmitter release during homeostatic plasticity.

Disclosures: Y. Cai: None. **T. Wang:** None. **J. Chen:** None. **A. Chakraborty:** None. **T. Cui:** None. **T. Wang:** None.

Poster

PSTR125. Transcription and Translation in Plasticity: New Tools and Novel Mechanisms

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR125.01/C30

Topic: B.05. Synaptic Plasticity

Support: NIH Grant AG068423
Alzheimer's Association Research Grant
WSU UROP Grant

Title: A Molecular Tool Designed For Selective Inhibition of Axonal Translation

Authors: *B. BONE, A. GRETZINGER, J. PARK;
Pharmacol., Wayne State Univ., Detroit, MI

Abstract: Protein synthesis is animportant cellular process to maintain cellular structure and functions.Neuronal morphology, characterized by lengthy axonal and dendritic projections,and activity-dependent plasticity are likely to encounter difficulties inpreserving synaptic proteins when and where they are needed. Evidence indicatespools of ribosomes and mRNAs that localize far from cell bodies, particularlyin the presynaptic terminals. However, the functional roles of presynaptictranslation in neuronal functions and behaviors remain elusive due to the lackof molecular tools capable of selective control of ribosomal activity at thepresynaptic terminal. Here, we develop a molecular tool designed to suppress ribosomalactivities compartmentally in the presynaptic terminal (termed prePSI) byutilizing a genetically encodable protein synthesis inhibitor (gePSI) and apresynaptic protein, Synaptophysin (Synph). We validated the efficacy of prePSIin translational inhibition in a heterologous cell system (HEK293T cells) byconducting puromycylation assays, which evaluate nascent protein synthesis byincubating cells with 3 μM puromycin for 10 min and subsequent anti-puromycinimmunostaining. Our preliminary data show a significant reduction in nascentprotein synthesis in prePSI-expressing cells compared with a negative controlthat expresses GFP-fused Synph (Synph-GFP) (88.1% reduction, n = 34-58 cells, $p < 0.0001$, Kruskal-Wallis test). Wefurther validated that the inhibitory effect of prePSI is comparable to that ofgePSI (global translation inhibitor), to ensure the fusion with Synph does notalter the efficacy ($p = 0.3982$, Kruskal-Wallis test). We also tested theselectivity of prePSI's translation inhibition by injecting an adeno-associatedvirus (AAV) expressing prePSI into the hippocampal dentate gyrus (DG).Following the trisynaptic circuit of the hippocampus, prePSI's translationinhibition efficacy was validated at the presynaptic terminals connected to theCA3 pyramidal neurons by conducting in vivo puromycylation and subsequentanti-puromycin immunostaining. Our data show a significant reduction in nascentprotein synthesis in the stratum lucidum, the DG-CA3 connecting layer of thehippocampus, in prePSI-injected samples, compared with that of aSynph-GFP-expressing control (n = 16 randomly chosen images from n = 4 mice pergroup, $p < 0.05$, Mann-Whitney tests). However, there was no apparentdecrease in anti-puromycin signals in the cell bodies of DG expressing prePSIcompared with those with Synph-GFP, suggesting that

prePSI is a selectiveinhibitor of axonal translation. Finally, we evaluated changes in fearconditioning with prePSI (n = 5 mice per group, female and male, in a blindedmanner to AAVs). We found that the prePSI-expressing group shows a significantdecrease in contextual and cued memory compared with a control group expressingSynph-GFP ($p < 0.01$, Mann-Whitney tests). These data suggest that prePSI isa selective inhibitor of axonal translation and that hippocampal axonaltranslation in the stratum lucidum is required for the development of fearmemories.

Disclosures: B. Bone: None. **A. Gretzinger:** None. **J. Park:** None.

Poster

PSTR125. Transcription and Translation in Plasticity: New Tools and Novel Mechanisms

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR125.02/C31

Topic: B.05. Synaptic Plasticity

Support: EMBO Postdoctoral Fellowship ALT 238-2021
ERC Grant 743216

Title: Elucidating the synaptic ribosome interactome

Authors: *A. M. BOURKE, K. DESCH, S. MOTA, J. D. LANGER, E. M. SCHUMAN; Max Planck Inst. for Brain Res., Frankfurt am Main, Germany

Abstract: Cue-specific, customized remodeling of synaptic proteomes is essential for proper neuronal function. During brain development and plasticity, local protein synthesis is differentially regulated in individual synaptic compartments to control synapse formation and strength (Bernard et al., 2022; Hafner et al., 2019), however the repertoire of molecular mechanisms used is not well-understood. A promising mechanism for sculpting synapse-specific proteomes is selective translation of synaptic mRNAs by ‘specialized ribosomes’ – ribosomes with different subunit compositions or associated proteins. This is supported by recent findings of synaptic mRNA translation on 80S monosomes (Biever et al., 2020; Glock et al., 2021) and context-specific ribosome remodeling in axons and dendrites (Shigeoka et al., 2019; Fusco et al., 2021). To what extent are ribosomes localized at synaptic compartments ‘special’, more dynamic or otherwise distinct from ribosomes found elsewhere in the cell? And can these properties be altered by synaptic plasticity? To begin to address these questions, we combined compartment-specific proximity labeling, sucrose cushion-based ribosome enrichment, and quantitative proteomics to map the protein interactomes of ribosomes in the nucleus (nuclear membrane), cytoplasm and dendritic spines of cultured rat hippocampal neurons. Candidate interactors, including several synaptic proteins, were validated by co-immunoprecipitation (co-IP) of endogenous ribosomes followed by Western blot and mass spectrometry (MS). Using co-IP/MS and enhanced-resolution image scanning microscopy, we are assessing the spatiotemporal profiles of these interactions following different plasticity paradigms. Preliminary results suggest

a dynamic interplay between ribosomes, synaptic proteins and actin-associated proteins, thereby offering insight into how synaptic ribosomes are ‘special’ and, more broadly, how precise spatial targeting of the translational machinery is achieved.

Disclosures: **A.M. Bourke:** None. **K. Desch:** None. **S. Mota:** None. **J.D. Langer:** None. **E.M. Schuman:** None.

Poster

PSTR125. Transcription and Translation in Plasticity: New Tools and Novel Mechanisms

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR125.03/C32

Topic: B.05. Synaptic Plasticity

Support: ERC Grant 743216
ERC Grant 101054512

Title: Transcriptomic characterization of synapse types and states

Authors: *M. JÜNGLING, J. PEREZ, S. TOM DIECK, M. VAN OOSTRUM, E. CIIRDAEVA, G. TUSHEV, N. FÜRST, E. SCHUMAN;
Max Planck Inst. for Brain Res., Frankfurt, Germany

Abstract: Neurons are remarkably complex cells. Their intricate morphology and the demands of synaptic plasticity pose unique challenges for synaptic proteostasis and proteome remodeling. A critical mechanism in neurons to meet these demands is mRNA transport from the soma to neuronal processes and subsequent local protein synthesis near or at synapses. Recent technical advances have pushed the transcriptomic characterization of neuronal subcellular compartments with neurotransmitter type specificity and under different plasticity contexts. However, a comprehensive characterization of synapse type- and state-specific transcriptomes is still lacking. To address this, we use mouse lines with presynaptic terminals that are fluorescently tagged in a cell-type specific manner. We then dissect the brain region of interest, homogenize the tissue and prepare synaptosomes - isolated presynaptic terminals often attached to the postsynaptic compartment - from adult animals of both sexes. We use Fluorescence-Activated Synaptosome Sorting to obtain millions of purified synaptosomes generated from specific cell types. This approach allows us to subsequently perform RNA sequencing, providing unprecedented insight into the transcriptomic diversity of synapse types. Profiling glutamatergic and GABAergic synaptosomes revealed that our sorting procedure successfully allows us to deplete non-synaptic genes originating from non-synaptosomal subcellular particles and free-floating RNA. Furthermore, our data confirm that glutamatergic and GABAergic synapses are transcriptomically distinct, with hundreds of genes uniquely enriched in each type. These genes include cell type markers for glutamatergic and GABAergic neurons like Camk2a and Parvalbumin, respectively. In a second line of experiments, we have established a complementary primary neuronal cell culture system, allowing us to profile transcriptome

remodeling in synapses after pharmacological manipulations. Our data suggest that the synaptic transcriptome also reflects different activity states with dozens of genes - many of which are immediate early genes - upregulated during homeostatic downscaling in glutamatergic, but not GABAergic synapses. Our approach reveals new insights into the transcriptomic diversity of synapse types and states. Also, it can be flexibly adapted to profile synapse remodeling during any form of plasticity or after any behavioral manipulation.

Disclosures: **M. Jüngling:** None. **J. Perez:** None. **S. tom Dieck:** None. **M. van Oostrum:** None. **E. Ciirdaeva:** None. **G. Tushev:** None. **N. Fürst:** None. **E. Schuman:** None.

Poster

PSTR125. Transcription and Translation in Plasticity: New Tools and Novel Mechanisms

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR125.04/C33

Topic: B.05. Synaptic Plasticity

Support: NSF NeuroNex 2014862

Title: In vivo visualization of local protein synthesis dynamics in the mouse cortex

Authors: *T. SPANÒ[1], P. G. DONLIN-ASP[3], G. TUSHEV[2], E. CIIRDAEVA[4], J. J. LETZKUS[6], E. M. SCHUMAN[5];
[1]Max Planck Inst. For Brain Res., Frankfurt Am Main, Germany; [2]Max Planck Inst. For Brain Res., Max Planck Inst. For Brain Res., Frankfurt, Germany; [3]the Univ. of Edinburgh, Edinburgh, United Kingdom; [4]Max Planck Inst. for Brain Res., Frankfurt am Main, Germany; [5]Erin M Schuman, Phd, Max Planck Inst. for Brain Res., Frankfurt/Main, Germany; [6]Inst. for Physiol., Univ. of Freiburg, Freiburg, Germany

Abstract: Synaptic plasticity is a key process for brain function and memory formation. This depends on the ability of neurons to remodel their synaptic proteomes with high temporal and spatial resolution, promptly delivering and degrading the right proteins at the correct locations. Decades of work have shown that local protein synthesis at synapses is one of the mechanisms that remodels the local proteome. *In vitro* and *ex vivo* studies have shown that neurons locally translate thousands of mRNAs, and that this process is required for several forms of long-term synaptic plasticity. In order to understand the role of local translation in physiological brain function and learning, one must study this phenomenon *in vivo*, in an intact neuronal circuit. Here, we present our efforts to visualize directly the protein synthesis dynamics from mRNAs in awake mice. We focus on cortical layer 1, a structure critical for learning that is devoid of neuronal cell bodies but enriched in their dendrites and axons. We show that layer 1 contains more than 2600 neuronal transcripts and exhibits ongoing protein synthesis. In order to visualize local translation in layer 1 dynamically, we created translation reporters by fusing the mRNA encoding a fast-folding fluorescent protein to the 3'untranslated regions of two synaptic transcripts, *Beta actin* and *Psd95*. We sparsely express these reporters in layer 5 excitatory

neurons and image their dendrites in layer 1, approximately 400 um away from their cell bodies. Using 2-photon imaging and fluorescence recovery after photobleaching (FRAP), we detect a rapid (protein synthesis dependent) emergence of reporter fluorescence in layer 1 dendrites. With this novel toolset, for the first time, we visualize local protein synthesis in intact brains in awake mice and are in a position to study its functional relevance in future studies.

Disclosures: **T. Spanò:** None. **P.G. Donlin-Asp:** None. **G. Tushev:** None. **E. Ciirdaeva:** None. **J.J. Letzkus:** None. **E.M. Schuman:** None.

Poster

PSTR125. Transcription and Translation in Plasticity: New Tools and Novel Mechanisms

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR125.05/C34

Topic: B.05. Synaptic Plasticity

Title: Exploring subcellular diversity in neuronal circuits through advanced transcriptomic approaches to study compartment-specific RNA profiles

Authors: ***J. PEREZ**[1], M. JÜNGLING[1], K. VERDAASDONK[2], Z. XIONG[2], M. VAN OOSTRUM[3], S. TOM DIECK[3], C. JOURDAN[2], R. ADRIAN[2], N. FUERST[2], G. TUSHEV[3], E. CIIRDAEVA[3], E. SCHUMAN[4];
[1]Max Planck for Brain Res., Frankfurt, Germany; [2]Synaptic Plasticity, Max Planck Inst. for Brain Res., Frankfurt am Main, Germany; [3]Synaptic Plasticity, Max Planck Inst. for Brain Res., Frankfurt, Germany; [4]Synaptic Plasticity, Erin M Schuman, Phd, Frankfurt/Main, Germany

Abstract: Neurons have complex structures (axons and dendrites) that create smaller compartments within them to sense, process, and transmit information semi-independently. It is not well understood how different neurons within a circuit represent diverse information across these smaller units, such as pre- and post-synaptic compartments, dendritic and axonal segments, and cell bodies and nuclei. This representation likely depends on the molecular differences between these units and their changing functional states. Our research aims to uncover the diversity within neuronal circuits by studying localized RNAs that enable changes in gene expression in space and time. We are developing advanced methods for isolating single subcellular units and detecting the small amounts of RNAs within them. We previously used laser capture microdissection to separate compartments from cultured rat hippocampal neurons, and single-cell RNA sequencing (scRNA-seq) to examine the local RNA content. We found that, like their excitatory counterparts, interneurons contain a rich repertoire of ~4000 dendritic mRNAs. To attain a higher level of resolution, we have developed SynDrops, a droplet microfluidic approach for characterizing the transcriptomic composition of individual synapses. This method involves sorting and purifying fluorescently labeled synaptic particles in a cell-type-specific manner. We then encapsulate individual synaptic particles into picoliter volume droplets containing the necessary components to convert synaptic RNAs to cDNA, adding a unique molecular barcode to the RNAs within each droplet. After sequencing the mixed contents of

these droplets, we use bioinformatics to reconstruct the RNA content of individual synapses based on their molecular barcodes. Following dimensionality reduction, several distinct synapse types and/or states are identified. Overall, our subcellular RNA analysis techniques will help us better understand the unique characteristics and functions of individual subcellular compartments within neuronal circuits.

Disclosures: **J. Perez:** None. **M. Jüngling:** None. **K. Verdaasdonk:** None. **Z. Xiong:** None. **M. van Oostrum:** None. **S. tom Dieck:** None. **C. Jourdan:** None. **R. Adrian:** None. **N. Fuerst:** None. **G. Tushev:** None. **E. Ciirdaeva:** None. **E. Schuman:** None.

Poster

PSTR125. Transcription and Translation in Plasticity: New Tools and Novel Mechanisms

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR125.06/C35

Topic: B.05. Synaptic Plasticity

Support: 2R37 NS036715-23

Title: Single-cell long-read sequencing reveals learning-specific alternative splicing in the mouse hippocampus

Authors: *S. CAVALIER, P. HOOK, W. TIMP, R. HUGANIR;
Johns Hopkins Sch. of Med., Baltimore, MD

Abstract: Activity-induced transcription enables learning and memory consolidation. This response is **cell-type specific** and involves thousands of genes. Despite targeted studies of specific genes emphasizing the functional importance of **alternative splicing** in this response, single-cell isoform quantification has been limited by the high input requirements (Pacific Biosciences) and low base calling accuracy (Nanopore) of long-read sequencers. Using a modified version of a Nanopore error-correction protocol developed by Volden et. al, 2018, we can sequence full-length barcoded cDNA from 10X Genomics while overcoming the error rates in Nanopore base calling that would otherwise occlude barcode demultiplexing and cell-type classification. We generate enough reads per cell (avg 2500 UMIs/cell) to confidently classify cell type and with an **average length of 900bp**. The complete exon coverage achieved by long-read sequencing enables splice-junction inferencing, isoform identification and novel isoform discovery at single-cell resolution. We applied this methodology to profiling the activity-induced transcriptome in the adult mouse hippocampus following **contextual fear-conditioning**, during which mice learn to associate a context with an aversive stimulus. We prepared libraries from wildtype mice that were exposed to contextual cues in either the presence (fear conditioned) or absence (unconditioned) of foot shocks and at one of two time points following exposure (15m, 60m). Libraries were also prepared from naïve, non-exposed animals. We used linear modeling software DeSeq2 to resolve genes and isoforms that are characteristic of each condition and time. We find that, across cell types, gene-level variance is sufficient to differentiate between naïve

and context-exposed cells; however, *isoform-level* variance better characterizes the transcriptional differences between experiencing a context (unconditioned) and learning something about it (fear conditioned). **We present the first known single-cell RNA-seq dataset to profile the activity-induced transcript isoform landscape in the mammalian brain.** Furthermore, we observe many genes with various functional annotations that have learning-specific transcript isoform expression, including chromatin remodelers, transcription factors, and synaptic proteins. We believe that this dataset will be foundational for understanding the function and regulation of learning-specific alternative splicing, possibly even elucidating some of the epigenetic substrates of learning as we continue with future directions.

Disclosures: S. Cavalier: None. **P. Hook:** None. **W. Timp:** None. **R. Huganir:** None.

Poster

PSTR125. Transcription and Translation in Plasticity: New Tools and Novel Mechanisms

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR125.07/C36

Topic: B.05. Synaptic Plasticity

Support: NIH T32 GM142519
Corrine F. Resta Endowed Doctoral Student Award
NIH AG068423

Title: Establishing a molecular tool to study dendritic protein synthesis in vivo

Authors: *A. GRETZINGER, J. PARK;
Wayne State Univ., Detroit, MI

Abstract: Protein synthesis is critical for learning and memory. The high degree of neuronal morphology and its activity-dependent plasticity are likely to face challenges in maintaining synaptic proteins appropriately. Evidence indicates the presence of synapse-associated ribosomes in dendritic spines and local mRNA pools being translated by ribosomes in neurites. In fact, several key proteins involved in maintaining the dendritic spine architecture are reported to be preferentially translated in the neuropil. Additionally, dysfunctional spine architecture is observed in neurodegenerative disorders such as Alzheimer's Disease (AD), indicating a possible disturbance in dendritic protein homeostasis. Despite the accumulating evidence supporting local translation, its roles in learning and memory remain elusive. To address this question, we generated a viral construct expressing a genetically encodable protein synthesis inhibitor designed to be targeted selectively to postsynaptic regions (postPSI). We validated the efficacy of our construct in a heterologous cell system utilizing a puromycylation assay. We then validated its compartment-specific inhibition of protein synthesis utilizing *in vivo* puromycylation assay in mice. Using this molecular tool, we evaluated the effect of dendritic protein synthesis on the spine plasticity of primary hippocampal neurons upon glutamate uncaging. Utilizing postPSI, we further investigated the role of dendritic protein synthesis in the

formation and maintenance of fear memories. These data may provide insights into the memory process mechanisms, which are of benefit in understanding the progression of neurodegenerative disorders.

Disclosures: A. Gretzinger: None. **J. Park:** None.

Poster

PSTR125. Transcription and Translation in Plasticity: New Tools and Novel Mechanisms

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR125.08/C37

Topic: B.05. Synaptic Plasticity

Support: NINDS Grant:1F31NS132558-01 (AMC)
Hope For Depression Research Foundation (EJN)
NIMH Grant: R01MH129306 (EJN)
NIMH Grant: R01MH116900 (IM)
HHMI Young Investigator Award (IM)

Title: Functional Role for Histone Serotonylation During Critical Periods of Postnatal Brain Development

Authors: *A. M. CUNNINGHAM[1], J. CHAN[2], M. S. ESTILL[2], E. BRINDLEY[2], L. SHEN[2], E. J. NESTLER[2], I. MAZE[2,3];
[1]Icahn Sch. of Med. At Mount Sinai Grad. Training Program In Neurosci., New York, NY; [2]Icahn Sch. of Med. At Mount Sinai, New York, NY; [3]Howard Hughes Med. Inst., New York, NY

Abstract: The serotonergic (5HTergic) system is implicated in a wide range of neurodevelopmental and neuropsychiatric phenomena. While 5HT actions have been assumed to work exclusively through 5HT receptors and their synaptic effects, recent studies established that 5HT forms covalent bonds with histone H3 glutamine 5 (H3 serotonylation). H3 serotonylation regulates normal patterns of neuroplasticity. However, functional roles for H3 serotonylation during post-natal neurodevelopment, a critical period of neuroplasticity, have largely been unexplored, and the impact of environmental stimuli (aberrant or otherwise) on this modification during early life remains unknown. Interestingly, we identified region- and developmental-dependent abundance of H3 serotonylation. We used FANS-coupled CUT&RUN to profile the cell type-specific developmental landscape of the mouse prefrontal cortex (PFC) a brain region that receives reciprocal projections from the DRN, which influences cortical 5HT release. We found that at postnatal days 10, 21, and 70, there was cell-type specific genomic distribution of H3 serotonylation in the male and female PFC. Importantly, we find that exposure to early life stress perturbs normal developmental trajectories of genomic enrichment of H3 serotonylation in a cell type-specific manner. Overall, we provide novel insight into how H3 serotonylation

regulates neurodevelopment and the mechanisms by which disruptions to this post-translational modification cause aberrant pathophysiological states.

Disclosures: **A.M. Cunningham:** None. **J. Chan:** None. **M.S. Estill:** None. **E. Brindley:** None. **L. Shen:** None. **E.J. Nestler:** None. **I. Maze:** None.

Poster

PSTR125. Transcription and Translation in Plasticity: New Tools and Novel Mechanisms

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR125.09/C38

Topic: B.05. Synaptic Plasticity

Support: NIH 5R01MH116900-03
HHMI Ian Maze

Title: Histone serotonylation dynamics in the context of mood disorders

Authors: *G. DI SALVO[1,2], A. AL-KACHAK[1], S. FULTON[3], J. CHAN[3], L. FARRELLY[3], I. MAZE[3];
[1]Neurosci., Icahn Sch. of Med. At MSSM Mount Sinai Grad. Training Program In Neurosci., New York, NY; [2]Maastricht Univ., Maastricht, Netherlands; [3]Icahn Sch. of Med. At Mount Sinai, New York, NY

Abstract: Histone proteins and their associated posttranslational modifications (PTMs) have been implicated in several pathologies. Recently, our lab demonstrated that in neuronal populations serotonin can be transamidated to glutamine 5 of histone H3, usually found alongside lysine 4 trimethylation, uncovering a novel combinatorial histone PTM named histone serotonylation. Although the histone serotonylation mark (H3K4me3Q5ser) has been linked to neuronal plasticity, little is known about how pathological states might affect its dynamics. Thus, we aimed to examine the role of H3K4me3Q5ser in the context of stress-induced depressive-like behaviors within the dorsal raphe nucleus (DRN) and the prefrontal cortex (PFC), regions involved in conditions associated to monoaminergic dysfunction.To this aim, we employed a chronic social defeat stress (CSDS) paradigm followed by 30 days of antidepressant (AD) administration in adult male mice. To assess whether changes in social interaction were correlated with potential chromatin-related changes, H3K4me3Q5ser levels were measured using western blots 24 hours and 30 days after the CSDS in the DRN. We found that after chronic exposure to stress, H3K4me3Q5ser displayed aberrant accumulation in DRN of susceptible mice, an effect that was not observed in stress-resilient mice and was reversed in response to 30 days of AD treatments. Additionally, we performed bulk RNA-seq and ChIP-seq to determine if H3K4me3Q5ser changes were upstream of DRN transcriptional plasticity. To further investigate the link between stress-mediated transcriptional and behavioral plasticity and aberrant histone serotonylation we employed a dominant negative viral vector approach to reduce levels of H3K4me3Q5ser in the DRN. Such approach reversed chronic stress-induced gene expression

programs and promoted behavioral resilience to stressful stimuli. Current research is now investigating these results in DRN's projection areas, such as the PFC.Finally, preliminary data from our lab suggested that histone serotonylation might occur in non-neuronal cells residing in the brain, such as glial cells. Given that perturbation in glial cell activity is thought to contribute to behavioral changes associated with depressive-like behavior, ongoing research will investigate the contribution of histone serotonylation changes in selected non-neuronal cell-types in the development of stress-induced depressive-like behaviors in mice. With a focus on abnormal H3K4me3Q5ser dynamics, these findings indicate a neurotransmission-independent role for 5-HT in stress-mediated transcriptional and behavioral plasticity.

Disclosures: G. Di Salvo: None. **A. Al-Kachak:** None. **S. Fulton:** None. **J. Chan:** None. **L. Farrelly:** None. **I. Maze:** None.

Poster

PSTR125. Transcription and Translation in Plasticity: New Tools and Novel Mechanisms

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR125.10/C39

Topic: B.05. Synaptic Plasticity

Support: R01 DA056595

Title: Identification of N-ribosyldihydronicotinamide: quinone reductase 2 (NQO2) as a novel reader of histone H3 serotonylation that contributes to neural gene expression

Authors: *M. CHEN[1], I. MAZE[1], H. LI[2];
[1]Icahn Sch. of Med. at Mount Sinai, New York, NY; [2]Tsinghua Univ., Beijing, China

Abstract: Identification of N-ribosyldihydronicotinamide: quinone reductase 2 (NQO2) as a novel reader of histone H3 serotonylation that contributes to neural gene expression
Min Chen[1], Lingchun Kong[1], Haitao Li[2], Ian Maze[1]
Monoaminergic neurotransmission in the central nervous system (CNS) plays a critical role in brain development and function, with alterations in monoamine production/signaling implicated in the development and treatment of many neurological diseases, including substance use disorders, mood syndromes and neurodegeneration. Serotonin – as well as other monoamines – had previously been shown to form covalent bonds with certain cytoplasmic proteins, catalyzed by the Transglutaminase 2 enzyme, and our group recently identified histone proteins as robust substrates for monoaminylation in brain (specifically histone H3 at position glutamine 5 – H3Q5ser). Our data indicate that histone serotonylation acts to alter the binding of histone/DNA modification interacting proteins and plays direct roles in neuronal transcription, particularly during periods of increased cellular activity. Furthermore, we have uncovered pathophysiological associations between altered levels of H3 monoaminylations and behavioral deficits observed in rodent models of disease. Importantly, however, the field has yet to uncover H3Q5ser specific 'readers' that may contribute to transcriptional plasticity in brain. As such, we recently profiled

H3Q5ser interacting proteins in cellular nuclear extracts via immunoprecipitations using biotinylated chemically modified H3 peptides, followed by LC-MS/MS mass spectrometric identification. In doing so, we found N-ribosyldihydronicotinamide: quinone reductase 2 (NQO2) to be robustly increased in its binding to H3 in the presence of Q5ser, an interaction that was subsequently validated using both biophysical and X-ray crystallography-based approaches. Then we characterized two endogenous mutants NQO2-I129EI129R and NQO2-I129R195E, which abolish the Q5ser binding ability, then followed by LC-MS/MS of Flag-HA tagged NQO2 and mutants, we identified NQO2 specifically interacting protein, OSBP2 (Oxysterol Binding Protein 2). Now, employing a wide variety of biochemical, genome-wide and protein engineering strategies, we are assessing functional roles for NQO2-H3Q5ser interactions in the regulation of permissive gene expression in neurons. In sum, our work has identified a previously uncharacterized, *bona fide* 'reader' of novel H3Q5ser, which likely contributes significantly to H3 serotonylation mediated gene expression in the CNS.

Disclosures: M. Chen: None. **I. Maze:** None. **H. Li:** None.

Poster

PSTR125. Transcription and Translation in Plasticity: New Tools and Novel Mechanisms

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR125.11/C40

Topic: B.05. Synaptic Plasticity

Support: 1R01HD097088-01A1
HHMI Ian Maze

Title: Role of Brwd1 in activity dependent regulation of gene expression in the Ts65Dn mouse model in Down syndrome

Authors: *S. DUTTA[1], E. BRINDLEY[1], S. FULTON[2], B. CETIN[1], I. MAZE[1];
[1]Icahn Sch. of Med. at Mount Sinai, New York, NY; [2]Columbia Univ., New York City, NY

Abstract: Down Syndrome (DS), caused by triplication of all or part of chromosome 21 (HSA21) in humans, is one of the most prevalent chromosomal abnormality disorders and is characterized by cognitive impairments and synaptic deficits among others. Previous studies, in both human and rodent models, have suggested that DS phenotypes are not driven by a 1:1 relationship between gene dosage and gene expression at trisomic loci. Instead, they may be driven by more complex mechanisms regulatory mechanisms accounting for the variability in the type and the severity of these clinical features. Epigenetic processes may also contribute to global patterns of transcriptional dysregulation, both during neurodevelopment and in adulthood. Our lab recently demonstrated that Brwd1, a WD-repeat and bromodomain-containing protein, encoded by HSA21, is upregulated in both trisomic human neurons and Ts65Dn mouse brain. We observed that Brwd1 triplication contributes to gene expression abnormalities in DS-like brain. Additionally, we found that DS-related physiological and hippocampal- dependent

cognitive impairments, in adult trisomic mice were rescued by the selective restoration of Brwd1 copy number. These data demonstrate a critical role of Brwd1 gene dosage in regulating aberrant neuronal gene expression patterns in DS-like brain, which precipitate synaptic and cognitive deficits associated with this disorder. To get a better understanding of the role that Brwd1 plays in activity- dependent regulation of neuronal impairments observed in DS- like rodent brains, specifically within the dorsal hippocampus, we have performed single nuclear RNA (snRNA)sequencing. We exposed male trisomic (*Ts65Dn*), WT, and Brwd1-rescue (*Ts65Dn; Brwd1+/-*) to a single foot shock of 0.8 mA and collected brains at 60 minutes. Bilateral punches of the dorsal hippocampal tissues were collected and processed for snRNA sequencing. We aim to a) identify specific hippocampal cell population which might contribute to learning and memory related deficits observed in DS-like mice; and subsequently c) target the identified cell populations to rescue the DS associated deficits. We also found that Brwd1 exerts its effects on synaptic plasticity, cognition, and gene expression in DS like brain by interacting with the BAF complex. To investigate this further, we are purifying its reader domains to understand how it interacts with Histone post translational modifications to guide BAF recruitment to neuronal chromatin, and how this process may be disrupted when triplicated in DS. Overall, these studies indicate Brwd1 is a key regulator of neuronal gene expression patterns in DS like brain.

Disclosures: S. Dutta: None. **E. Brindley:** None. **S. Fulton:** None. **B. Cetin:** None. **I. Maze:** None.

Poster

PSTR125. Transcription and Translation in Plasticity: New Tools and Novel Mechanisms

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR125.12/C41

Topic: B.05. Synaptic Plasticity

Support: NIH Grant MH126534
NIH Grant DA056595
NIH Grant MH116900
NIH Grant HD097088

Title: Cellular mechanisms contributing to the persistent effects of pregnancy and postpartum experiences on brain function

Authors: *J. CHAN, G. DI SALVO, A. CUNNINGHAM, E. BRINDLEY, S. DUTTA, I. MAZE;
Mount Sinai Sch. of Med., New York, NY

Abstract: Pregnancy represents an incredible physiological stressor, yet while the effects of environmental and psychological stresses are well-documented, how reproductive experiences produce lasting impact on the maternal brain remain unknown. Moreover, prior pregnancies (parity) are risk factors for several disorders including perinatal/postpartum mood and affective

disorders and Alzheimer's Disease, yet not all individuals who experience pregnancy and childbirth develop brain disorders later in life. Therefore, there is great need to understand the biological processes that both orchestrate and disrupt parity effects in the brain to promote later disease susceptibility. Using mouse models, our lab identified the dorsal hippocampus (dHpc) as a brain region with robust transcriptional alterations across the gestational and postpartum periods and up to 1 month following offspring weaning. These transcriptional changes suggest altered neuronal and glial composition compared to nulliparous females, and were associated with improved performance on context-dependent fear conditioning, a behavioral task reliant on dHpc function. To determine the specific contributions of pregnancy and pup interactions on parity programming of the dHpc, we performed RNA-sequencing on pregnancy only vs. pup sensitized females. We show that while pregnancy is the major contributor of neuroplasticity changes, complete primiparous programming of the dHpc involves additional postpartum exposures. Thus, we next tested whether disruptions over the postpartum period could impact primiparity-dependent changes. We utilized a model of maternal separation stress that incorporated limited nesting and daily pup separations over ten days. Our results show that this postpartum stress model inhibited both the transcriptional changes and behavioral improvements observed in control primiparous dams. To comprehensively define the impact of parity vs. postpartum stress effects, we performed single nuclei RNA-sequencing on the mouse dHpc. Analysis of neuronal subclusters revealed pregnancy-specific effects in select interneuron populations that are prevented by postpartum stress. Ongoing work will test the contribution of these interneuron-specific changes to primiparous-dependent behavioral adaptations. Overall, we suggest pregnancy and postpartum exposures elicit long-term cognitive improvements, which are vulnerable to disruptions by postpartum stress. These studies provide insight into the molecular mechanisms contributing to long-term effects of parity on the brain, and the environmental triggers that may interact with them to influence brain health.

Disclosures: **J. Chan:** None. **G. Di Salvo:** None. **A. Cunningham:** None. **E. Brindley:** None. **S. Dutta:** None. **I. Maze:** None.

Poster

PSTR125. Transcription and Translation in Plasticity: New Tools and Novel Mechanisms

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR125.13/C42

Topic: B.05. Synaptic Plasticity

Support: Ministerio de Ciencia e Innovación SAF2016-80027-R
Ministerio de Ciencia e Innovación PID2019-106615RB-I00
Ministerio de Ciencia e Innovación PDC2021-121350-I00
Instituto de Salud Carlos III, CIBERNED CB06/05/0042
APD is supported by a postdoctoral Juan de la Cierva-Incorporación fellowship IJC2019-042468-I
AdB is supported by a Personal Investigador en Formació fellowship FI_B00858 (AGAUR-Generalitat de Catalunya)

AD is supported by a Formación de Profesorado Universitario fellowship FPU 18/02486

Title: CREB/CRTC1 regulate a conserved neuronal activity-dependent gene program mediating synapse-to-nucleus signaling and synaptic plasticity

Authors: *A. J. PARRA-DAMAS[1,2], A. DEL SER-BADIA[1,2], A. DEPRADA[1,2], J. RODRÍGUEZ-ALVAREZ[1,2], C. A. SAURA[1,2];
[1]Inst. of Neurosci., Univ. Autònoma de Barcelona, Bellaterra, Spain; [2]Ctr. de Investigación Biomédica en Red Enfermedades Neurodegenerativas (CIBERNED), Madrid, Spain

Abstract: Long-term synaptic plasticity and memory requires neuronal activity-induced gene expression regulated by the transcription factor cAMP-response element binding protein (CREB). CREB-dependent gene expression requires transcriptional coactivators, including CREB binding protein (CBP/P300) and CREB-regulated transcription coactivator-1 (CRTC1). However, the specific gene programs regulated by CREB/CRTC1 during induction of neuronal activity remain largely unknown. Here, we used chromatin immunoprecipitation sequencing (ChIP-seq) to analyze the genome-wide occupancy profiles of CREB and CRTC1 in primary mouse cultured neurons in both basal and stimulated conditions after treatment with forskolin and potassium chloride (FSK/KCl), which induces CREB phosphorylation as well as CRTC1 dephosphorylation, nuclear translocation and binding to promoter regions of target genes. We detected strong induction of CRTC1 occupancy in CREB target genes upon FSK/KCl treatment, including proximal promoter regions and distal regions comprising well-characterized neuronal activity-regulated enhancers. Interestingly, we identified activity-dependent binding of CREB/CRTC1 at genes mediating neuronal excitability and synaptic plasticity, including neurotransmitter receptors, synaptic proteins, and transcriptional regulators involved in synapse-to-nucleus signaling. Importantly, analysis of an independent CREB/CRTC1 ChIP-seq dataset generated from human iPSC-derived neuronal cells revealed that these binding regions are conserved in mouse and human orthologs. Moreover, GO enrichment and machine learning-based protein-protein interaction (PPI) network analyses suggest that genes mediating synapse-to-nucleus signaling (including most known synaptonuclear factors and direct interacting modulators) are collectively regulated by CREB/CRTC1, and that protein kinase C (PKC) is a key gene product regulating synaptic networks. In agreement with these in silico analyses, we show that CRTC1 mediates NMDA receptor transmission and synaptic potentiation in the hippocampus by regulating protein kinase C (PKC)-induced phosphorylation and synaptic recruitment of GluN1. At the structural and functional level, CRTC1 is essential for dendritic spine morphology and synaptic plasticity, including long-term potentiation and depression, and long-term memory. In conclusion, CRTC1/CREB signaling controls a gene program supporting synapse-to-nucleus communication that is highly conserved between mice and humans and involves NMDAR-dependent synaptic plasticity through transcriptional and local synaptic mechanisms.

Disclosures: A.J. Parra-Damas: None. **A. del Ser-Badia:** None. **A. Deprada:** None. **J. Rodríguez-Alvarez:** None. **C.A. Saura:** None.

Poster

PSTR125. Transcription and Translation in Plasticity: New Tools and Novel Mechanisms

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR125.14/C43

Topic: B.05. Synaptic Plasticity

Support: R01EY011261 to HTC
R01EY027437 to HTC
P41GM103533 JRY
R01MH067880 JRY

Title: Cell-type specific proteomic analysis reveals an activity-induced protein synthesis inhibitor that regulates neuronal plasticity

Authors: *Y. XIE[1], R. WANG[1], D. MCCLATCHY[1], Y. MA[1], E. MCAULIFF[2], X. DONG[2], J. YATES, III[1], H. CLINE[1];
[1]Neurosci., Scripps Res., San Diego, CA; [2]UC San Diego, San Diego, CA

Abstract: Activity-induced protein dynamics is the underlying player of neuronal plasticity. A direct interrogation of activity-dependent proteome is a crucial step towards understanding protein components and their function in neuronal plasticity. Here we employed cell-type specific metabolic labeling and bio-orthogonal non-canonical amino acid tagging, to sample and detect cell-type specific nascent proteome upon visual experience in mouse visual cortex. We detected 2092 proteins in excitatory/inhibitory mouse neuron, at P28 and P56. We quantified both unique and shared proteins significantly regulated in these conditions. Correlation analysis of our proteomics with RNAseq and Riboseq only showed alignment to a limited extend, which indicates the involvement of regulatory elements at different levels. Pathway analysis revealed protein clusters for synaptogenesis, cytoskeleton remodeling and synaptic transmission. To further understand their functions, we designed a phenotypic screening using high-content imaging for 11 candidates in vitro. While all candidates showed significance in different aspects, one showed unexpected effect in suppressing global protein synthesis rate. Conversely, shRNA against this protein showed the opposite result. Moreover, different stimulants, both in vitro and in vivo, can increase its expression at protein level. The effect size is proportional to the intensity and duration of stimulation. During development, eye opening induces a surge in its expression, which persists throughout critical period and returns to low level by the closing of the critical period. With stimulation, the newly synthesized protein puncta visualized through FUNCAT-PLA were observed in nucleus, ER and neuronal processes, based on which we speculate that this protein regulates synthesis through various mechanisms. To examine its role in neuronal plasticity, we designed knockdown and overexpression experiments, where we found KD increases excitatory synapse density, but decreases spontaneous firing frequency. Conversely, OE decreases excitatory synapse density and increases inhibitory density and synchronous firing frequency. Therefore, we hypothesize that this protein candidate responds to activity by suppressing the synthesis of a selective group of protein, and as a result, weak synapses are eliminated to shape a more efficient network. In summary, we conducted a comprehensive interrogation of activity-induced protein dynamics in a cell-type specific manner, generated a

rich resource which leads to the discovery of an activity-dependent protein synthesis inhibitor with a function on refining neuronal network.

Disclosures: **Y. Xie:** None. **R. Wang:** None. **D. McClatchy:** None. **Y. Ma:** None. **E. McAuliff:** None. **X. Dong:** None. **J. Yates:** None. **H. Cline:** None.

Poster

PSTR125. Transcription and Translation in Plasticity: New Tools and Novel Mechanisms

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR125.15/C44

Topic: B.05. Synaptic Plasticity

Support: NIA/NIH R56AG072676

Title: Age-related alterations in the expression of neuronal regulators of synaptic plasticity in the mouse cortex

Authors: *C. SEMMEDI[1,3], R. MOSTANY[3,2];
[1]Sch. of Sci. and Engin., [2]Dept. of Pharmacology, Sch. of Med., Tulane Univ., New Orleans, LA; [3]Tulane Brain Inst., New Orleans, LA

Abstract: Aging, even in the absence of pathologies, is linked to cognitive deficits associated with impairments in synaptic plasticity. Previous studies from our laboratory have shown deficits in synaptic connectivity and responses to sensory-evoked plasticity in cortical pyramidal neurons of aged mice, implying impairments in synaptic plasticity in the aged cortex. This is also supported by studies showing that the expression of key promoters of synaptic plasticity, such as the immediate early gene Arc, brain-derived neurotrophic factor (BDNF), and tumor necrosis factor alpha (TNF-α) is impaired in the aged brain (Penner et al., 2011; Erickson et al., 2010; Porcher et al., 2021). In order to determine the genetic underpinnings of the neuronal defects seen in the aged cortex, we assessed the expression levels of these regulators of synaptic plasticity exclusively in neuronal populations of the brain cortex of mice. To achieve this, we utilized a papain dissociation system combined with a magnetically-activated cell sorting system (MACS) to selectively obtain cortical neurons of young (3- to 7-month-old) and aged (>21-month-old) mice. mRNA of each age group was collected to analyze the expression of Arc, BDNF, TNF-α, as well as the receptors for the latter two, tropomyosin receptor kinase B (TrkB), and tumor necrosis factor receptor 1 (TNFR1), respectively. To maximize the information that can be extracted from the limited RNA samples collected, droplet-digital PCR (ddPCR), an advanced PCR technology with higher accuracy and consistency than traditional PCR methods, was utilized for RNA quantification. Based on our findings, aged mice had reduced expression of Arc and BDNF, and increased expression of TNF in their cortical neurons, whereas expression of TrkB and TNFR1 was unaltered across age groups. Utilization of MACS followed by ddPCR allowed us to specifically examine neuronal expression of various genes in the cortex and

confirmed that neuronal expression of the key plasticity regulators studied is altered in the cortex with age.

Disclosures: C. Semmedi: None. **R. Mostany:** None.

Poster

PSTR125. Transcription and Translation in Plasticity: New Tools and Novel Mechanisms

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR125.16/C45

Topic: B.05. Synaptic Plasticity

Support: GRIEG 2019/34/H/NZ3/00733 (Norway Grants)

Title: The effect of TENT2 cytoplasmic poly(A) polymerase knock-in on electrical properties of neuronal membrane

Authors: *P. WARDASZKA[1], T. WOJTOWICZ[2], F. P. PAUZIN[3], C. R. BRAMHAM[3], M. DZIEMBOWSKA[1];
[1]Ctr. of New Technologies, Univ. of Warsaw, Warsaw, Poland; [2]Nencki Inst., Warsaw, Poland; [3]Dept. of Biomedicine, Univ. of Bergen, Bergen, Norway

Abstract: TENT2 (also called GLD2) is a cytoplasmic poly(A) polymerase. It catalyzes the process of polyadenylation, an elongation of poly(A) tails at the 3' ends of mRNAs. This posttranslational modification increases mRNA stability. Polyadenylation of mRNA molecules occurs mainly in the nucleus, before their transport to the cytoplasm. However, it can also take place in the cytoplasm of neurons. TENT2 is expressed in several brain structures including hippocampus of a mouse brain. We used an in-house generated Tent2 knock-in mice to study the effect of lack of Tent2 enzyme on neuronal function.We have analyzed passive membrane properties and membrane excitability of CA1 pyramidal neurons in acute hippocampal slices of wild-type and Tent2 knock-in mice. We found that Tent2 knock-in neurons exhibited increased rheobase, the minimal electrical current that is necessary to elicit an action potential compared to wild-type neurons (Mann-Whitney test, $p= 0.026$). In addition, the voltage-gated sodium channels (NaV) currents were measured. In Tent2 knock-in neurons the maximal value of NaV currents were reached at lower voltage (Mann-Whitney test, $p=0.032$). Next, we performed recordings of evoked field potentials in Sch-CA1 projection in acute hippocampal slices. In agreement with patch-clamp results, Tent2 knock-in mice exhibited 3-fold larger population spike amplitude relative to wildtype control (unpaired t test, $p<0.0001$). In contrast, no change in synaptically evoked population spike amplitude was observed in the perforant path-dentate gyrus (DG) projection *in vivo.* Finally, in order to determine the potential consequences of TENT2 deficit on synaptic transmission, patch clamp recordings of miniature excitatory postsynaptic currents (mEPSCs) were performed. mEPSCs were measured in the hippocampal primary culture. The cumulative distribution function of mEPSCs amplitudes was shifted toward larger values for Tent2 knock-in neurons (Kolmogorov-Smirnov test, $p<0.00001$) but the distributions

of inter-event time intervals remained similar. In conclusion, lack of TENT2 resulted in increased responsiveness of neuronal membrane to excitation, which is probably mediated by an altered functioning of voltage-gated Na+ channels. Probably this effect is region-specific as it was registered only in the Sch-CA1 projection. In addition, TENT2 deficit resulted in increased excitatory synaptic transmission most likely due to a higher number of AMPA/kainate receptors in postsynaptic membrane compared to wild-type neurons. Thus, cytoplasmic polyadenylation mediated by TENT2 may regulate the postsynaptic transmission and neuronal excitability.

Disclosures: P. Wardaszka: None. **T. Wojtowicz:** None. **F.P. Pauzin:** None. **C.R. Bramham:** None. **M. Dziembowska:** None.

Poster

PSTR125. Transcription and Translation in Plasticity: New Tools and Novel Mechanisms

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR125.17/C46

Topic: B.05. Synaptic Plasticity

Support: R01 NS057819-16

Title: Profiling neuronal activity-dependent molecular changes in a mouse model of Rett Syndrome

Authors: *Y. LI[1], J.-P. REVELLI[1], S.-R. WU[1], A. ANDERSON[1], J. ZHOU[1], H. ZOGHBI[1,2]; [1]Baylor Col. of Med., Houston, TX; [2]Howard Hughes Med. Inst., Houston, TX

Abstract: Rett syndrome (RTT) is a devastating neurological disorder caused by loss-of-function mutations in the X-linked gene *Methyl-CpG-binding protein 2* (*MECP2*). RTT occurs mostly in girls, who develop normally for the first 6-18 months and then experience profound, progressive motor and cognitive decline accompanied by breathing abnormalities. Through studies in patients and mouse models, we know that RTT is characterized by neuronal hypoactivity and altered expression of a broad range of neuronal genes. Activating RTT neurons through deep brain stimulation (DBS) enhanced neuronal activity and hippocampal plasticity and eventually improved learning and memory in RTT mice by normalizing the expression of many genes encoding synaptic proteins. Recently, we showed that repetitive behavioral training in RTT mice during the pre-symptomatic but not post-symptomatic stage can rescue certain behavioral abnormalities and improve the electrophysiological properties of neurons involved in those behavioral tasks (task-specific neurons). Interestingly, chemogenetic activation of task-specific neurons can achieve a similar behavioral rescue as that of repetitive training. Therefore, we hypothesize that pre-symptomatic training elicits transcriptional responses in the task-specific neurons which contribute to the behavioral improvement. To test this hypothesis, we combined FosTRAP technique with a conditional nuclear fluorescent label to isolate task-specific neurons during the behavioral training in both pre- and post-symptomatic RTT mice. In RTT mice carrying INTACT and fosTRAP alleles, we successfully showed early training mitigates learning

and memory deficits. Task-specific neurons are isolated and sequenced. Single-nucleus RNAseq of FosTRAPed neurons in pre- and post- symptomatic trained RTT and WT mice captured identical cell type, but differences were identified in cell type proportions. This will give us the opportunity to characterize the transcriptional signature of both wild-type and RTT neurons in response to physiological activity and allow us to compare the transcriptomic effect of pre- and post-symptomatic training on the task-specific neurons. Results from this study will shed light on our understanding of the key molecular drivers of RTT pathogenesis. Furthermore, studying the transcriptomic differences between pre- and post-symptomatic training models will allow us to parse apart the causative versus secondary changes in RTT and potentially help us identify specific molecular pathways that can be targeted for therapeutics.

Disclosures: **Y. Li:** None. **J. Revelli:** None. **S. Wu:** None. **A. Anderson:** None. **J. Zhou:** None. **H. Zoghbi:** None.

Poster

PSTR125. Transcription and Translation in Plasticity: New Tools and Novel Mechanisms

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR125.18/C47

Topic: B.05. Synaptic Plasticity

Support: German Research Foundation

Title: Cyclase-associated protein 1 (CAP1) represses MRTF-SRF-dependent gene expression in the mouse cerebral cortex

Authors: *S. KHUDAYBERDIEV[1], A. HEINZE[1], N. TRAUSCH[1], U. LINNE[2], M. RUST[1]; [1]Inst. für Physiologische Chemie, Philipps-Universitaet Marburg, Marburg, Germany; [2]Fachbereich-Chemie, Philipps-Universität Marburg, Marburg, Germany

Abstract: Serum response factor (SRF) is a ubiquitously expressed transcription factor essential for brain development and function. SRF activity is controlled by two competing classes of coactivators, myocardin-related transcription factors (MRTF) and ternary complex factors (TCF), which introduce specificity into gene expression programs. To date, only few brain studies investigated upstream regulatory mechanisms, which mainly focused on TCF. Since an inhibitory function of monomeric actin towards MRTF-SRF signaling is well-established, we hypothesized a regulatory role for the key actin regulator ADF/cofilin. Surprisingly, ADF/cofilin was largely dispensable for neuronal MRTF-SRF activity. Instead, reporter assays combined with pharmacological and genetic approaches in isolated mouse neurons identified cyclase-associated protein 1 (CAP1) as an important regulator of this pathway. CAP1 promotes cytosolic MRTF retention and represses neuronal MRTF-SRF signaling via an actin-dependent mechanism that requires two specific protein domains. Further, deep RNA sequencing and mass spectrometry in mutant mice proved CAP1's *in vivo* relevance for this pathway in the cerebral

cortex, and led to the identification of neuronal MRTF-SRF target genes. Together, we identified CAP1 as a novel and crucial repressor of neuronal MRTF-SRF signaling

Disclosures: **S. Khudayberdiev:** None. **A. Heinze:** None. **N. Trausch:** None. **U. Linne:** None. **M. Rust:** None.

Poster

PSTR125. Transcription and Translation in Plasticity: New Tools and Novel Mechanisms

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR125.19/C48

Topic: B.05. Synaptic Plasticity

Support: NIH Grant MH122961
NIH Grant NS083085
NIH Grant MH125772
NIH Grant MH116673
NRSA Fellowship MH109267

Title: Imaging single mRNAs provides insights into the mechanisms of long term protein maintenance required for memory

Authors: *S. DAS[1], N. MARTIN[1], D.-W. HWANG[1], P. J. LITUMA[1,2], P. E. CASTILLO[1], R. H. SINGER[1];
[1]Albert Einstein Col. of Med., Bronx, NY; [2]Weill Cornell Med., New York, NY

Abstract: Spatio-temporal control of gene expression in a neuronal network is an essential element of memory formation. One way of achieving precise gene control is by localizing mRNAs to synapses in an activity-dependent manner, and locally translating them to supply new proteins for long-term synaptic plasticity. However, the dynamics of individual mRNAs in response to stimulation and how they are regulated to achieve efficient spine remodeling remains unknown. We address these questions by high-resolution imaging of mRNAs and proteins in living neurons from cultures and ex vivo tissue. To this end, we generated knock-in mouse where endogenous *Arc* and *β-actin* genes are tagged in their 3'UTR with orthologous bacteriophage-derived stem loops that bind to different fluorescent coat proteins. By real-time imaging of both mRNAs in the same neuron, we demonstrated how these two plasticity-related mRNAs are regulated by distinct mechanisms to support long term spine remodeling. While constitutive, long-lived *β-actin* mRNAs persisted in the dendrites and undergo multiple rounds of translation, inducible *Arc* transcripts with short half-lives displayed transient localization with rapid degradation kinetics. These findings raised the conundrum of how unstable *Arc* mRNAs encoding short-lived proteins facilitate long-term physiological effects. Unexpectedly, we observed that the *Arc* gene but not *β-actin*, underwent cycles of transcriptional reactivation without any additional stimulation, to ensure an intermittent supply of mRNAs to stimulated spines over several hours. These cycles of transcription were coordinated with local translation

events to assemble a “hotspot” of Arc protein synthesis that replenished Arc levels over time to support long-lasting spine-remodeling. We propose that repetitive cycling of the gene provides a mechanism by which a short-lived event can be sustained, consistent with that necessary for long-term memory.

Disclosures: S. Das: None. **N. Martin:** None. **D. Hwang:** None. **P.J. Lituma:** None. **P.E. Castillo:** None. **R.H. Singer:** None.

Poster

PSTR125. Transcription and Translation in Plasticity: New Tools and Novel Mechanisms

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR125.20/C49

Topic: B.05. Synaptic Plasticity

Support: NIH Grant NS083085

Title: Visualizing the activity-dependent behavior of endogenous CaMKIIα transcripts at dendritic spines elucidates mRNA-mediated synaptic tagging mechanism

Authors: *D. HWANG, S. DAS, R. H. SINGER;
Cell Biol., Albert Einstein Col. of Med., Bronx, NY

Abstract: Calcium/calmodulin-dependent protein kinase type II (CaMKII) is a multimeric calcium-dependent kinase that serves to integrate transient calcium signals into long-term synaptic plasticity and structural changes at dendritic spines. Persisting phosphorylation among the subunits of CaMKII dodecamer has long been recognized as a key process that facilitates propagation of CaMKII-mediated signals in a self-contained manner. While an exchange of unphosphorylated CaMKII subunits with phosphorylated subunits of dodecameric holoenzymes has been postulated as the mechanism responsible for sustaining autophosphorylation of CaMKII, it remains unclear how an infusion of the protein subunits is achieved in each dendritic spine. We hypothesized that the major CaMKIIα subunits become available to stimulated dendritic spines via activity-dependent *de novo* local protein synthesis, which requires *Camk2a* mRNAs, encoding the CaMKIIα subunit, to localize to the corresponding spines and undergo rounds of on-site translation during synaptic changes. To test this hypothesis, we implemented single-molecule imaging approach to examine real-time behaviors of endogenous *Camk2a* mRNAs at dendritic spines. Live imaging of *Camk2a* mRNA molecules in hippocampal neurons revealed that upon stimulation, *Camk2a* transcripts localized to stimulated spines within ~1 minute, and this spine localization persisted up to 1 hour. While localized within the spines, mRNAs underwent *de novo* protein synthesis. Both spine localization and *de novo* protein synthesis were dependent on the activity of the N-methyl-D-aspartate receptors (NMDARs) during stimulation. Additionally, we have found that spine localization and *de novo* local protein synthesis of *Camk2a* mRNAs required the presence of cis regulatory elements in the 3’UTR, which are known binding elements for the interaction with the Cytoplasmic polyadenylation

element binding (Cpeb) proteins. Together, these findings suggest that activity-dependent localization and *de novo* protein synthesis of *Camk2a* mRNAs may serve to tag stimulated dendritic spines by supplying a spine-specific pool of CaMKIIα subunits during the maintenance phase of long-lasting synaptic changes.

Disclosures: D. Hwang: None. **S. Das:** None. **R.H. Singer:** None.

Poster

PSTR125. Transcription and Translation in Plasticity: New Tools and Novel Mechanisms

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR125.21/C50

Topic: B.05. Synaptic Plasticity

Support: NIH Grant MH129760
NIH Grant MH107254

Title: The P-TEFb inhibitory complex is correlated with human Alzheimer's disease pathology and regulates the rate that immediate early genes can be fully reinduced during repeated bouts of neuronal depolarization

Authors: Y. WU[1], S. HANSEN[1], T. J. HOHMAN[1], G. KAAS[2], R. J. COLBRAN[3], ***C. GREER**[4,5];
[1]Ctr. for Human Genet., [2]Genet. Med., Vanderbilt Univ. Med. Ctr., Nashville, TN; [3]Dept Molec Physiol & Biophysics, Vanderbilt Univ. Sch. Med., Nashville, TN; [4]Pharmacol., Vanderbilt Univ., Nashville, TN; [5]Mol. Pharmacol. and Neurosci., Loyola Univ. Chicago Hlth. Sci., Maywood, IL

Abstract: Rapid and transient induction of gene expression in neurons is necessary for the synaptic plasticity underlying learning and memory. Genes that are activated in neurons in response to stimuli, known as immediate early genes (IEGs), exist in a poised transcriptional state where RNA polymerase II (RNAP2) has initiated transcription, but pauses just downstream of the gene promoter. Neuronal depolarization releases the paused RNAP2 to complete the synthesis of the messenger RNA (mRNA) transcript. The release of poised RNAP2 requires the positive transcription elongation factor b (P-TEFb) protein heterodimer. In many cells, P-TEFb is sequestered into a larger inactive complex containing Hexamethylene bisacetamide inducible protein 1 (HEXIM1) under baseline conditions. Stimuli that induce IEG expression dissociate HEXIM1 from P-TEFb, allowing P-TEFb to release the paused RNAP2 and activate gene transcription. Inhibitors of elongation improve cognition in Alzheimer's disease (AD) model mice, and certain components of this inactive complex have been linked to clinical cases of human developmental disorders presenting with cognitive impairment. We find that the expression levels of mRNAs encoding components of the inactive P-TEFb complex, especially HEXIM1, are highly correlated with amyloid beta and neurofibrillary tangle pathologies as well as poorer cognition in several brain regions in bulk RNA sequencing data from human AD

postmortem tissue. Moreover, single nucleus RNA sequencing analysis revealed associations between neuronal HEXIM1 expression and AD pathology, further underscoring the importance of better understanding the regulation of P-TEFb in neurons. Depolarization of mouse primary neuronal cultures using potassium chloride stimulates IEG expression, and we now find that the transcriptional response to a second depolarizing stimulation is transiently suppressed for a period of hours after the first stimulation. During this transcriptional refractory period, HEXIM1 mRNA levels are induced, and HEXIM1 protein levels are reduced, suggesting that the neurons may be recovering to re-instate the poised baseline state where HEXIM1 is once again sequestered by P-TEFb. In support of this theory, we find blocking P-TEFb kinase activity during the initial stimulation prevents the suppression of the transcriptional response to a second stimulus. Together, our findings suggest that the HEXIM1/P-TEFb complex has an important role in setting and resetting the poised state that allows for the robust activation of genes necessary for synaptic plasticity, and that this regulation may be disrupted in diseases affecting memory.

Disclosures: **Y. Wu:** None. **S. Hansen:** None. **T.J. Hohman:** None. **G. Kaas:** None. **R.J. Colbran:** None. **C. Greer:** None.

Poster

PSTR125. Transcription and Translation in Plasticity: New Tools and Novel Mechanisms

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR125.22/C51

Topic: B.05. Synaptic Plasticity

Support: NIH R01-NS113600
NIH R01-DA17392
NIH R01-MH125772
NIH R01-MH116673
Junior Investigator Neuroscience Research Award
RFK-IDDRC Pilot Grant
American Epilepsy Society Postdoctoral Fellowship

Title: Postsynaptic protein synthesis is required for presynaptic LTP and is dysregulated in a mouse model of Fragile X Syndrome

Authors: *C. BERTHOUX[1], P. E. CASTILLO[1,2];
[1]Dominick P. Purpura Dept. of Neurosci., [2]Dept. of Psychiatry and Behavioral Sci., Albert Einstein Col. of Med., Bronx, NY

Abstract: Loss of Fragile X messenger ribonucleoprotein 1 (FMRP) causes Fragile X Syndrome (FXS), the leading monogenic cause of cognitive impairments. While FMRP is a mRNA translation repressor that regulates synaptic structure and function, how FMRP loss leads to synaptic and circuit defects in FXS remains poorly understood. In the dentate gyrus, a key brain

area involved in memory formation and epilepsy, granule cells (GCs) and hilar mossy cells (MCs) establish a recurrent GC-MC-GC excitatory loop. MC-GC synapses undergo NMDA receptor-independent and presynaptically-expressed form of long-term potentiation (MC-GC LTP) whose induction requires postsynaptic BDNF/TrkB signaling. By increasing the excitation/inhibition (E/I) balance, MC-GC LTP enhances GC output, thus activating the GC-MC-GC recurrent circuit and facilitating epileptic activity *in vivo*. Therefore, we hypothesized that abnormal strengthening of MC-GC transmission in FXS, by disrupting the E/I balance, significantly contributes to cognitive deficits and seizure susceptibility. We found that MC-GC synaptic strength is enhanced, and MC-GC LTP is occluded in *Fmr1* knockout mice, a model for FXS, and in conditional *Fmr1* knockout from GCs, but not MCs, suggesting a role for postsynaptic FMRP at MC-GC synapse. We then examined whether protein synthesis, which can be induced by BDNF/TrkB signaling cascade, is required for long-term changes at MC-GC synapses. Using puromycylation labeling to measure newly synthesized proteins, we found that MC-GC LTP is associated with increased puromycin signal in the GC layer but also in the inner molecular layer, strongly suggesting newly synthesized proteins at the MC-GC synapse. Accordingly, we showed that bath application of the protein synthesis inhibitors anisomycin or cycloheximide abolished MC-GC LTP, and loading GCs with the membrane impermeant protein synthesis inhibitor M7 also prevented LTP, indicating that this form of plasticity requires postsynaptic *de novo* protein synthesis. Finally, blocking protein synthesis during the induction, but not during the maintenance, prevented MC-GC LTP, suggesting the involvement of fast protein synthesis. Altogether, our findings indicate that dysregulated MC-GC synaptic function may be implicated in FXS.

Disclosures: **C. Berthoux:** None. **P.E. Castillo:** None.

Poster

PSTR125. Transcription and Translation in Plasticity: New Tools and Novel Mechanisms

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR125.23/C52

Topic: B.05. Synaptic Plasticity

Support: NRF-2020R1A5A1019023
NRF-2022R1A2C1004913
KHIDI-HU21C0071
BK21 Four Biomedical Science Program

Title: Regulate of local translation in response to neuronal stimuli in primary cultured neurons

Authors: *D.-H. PARK[1,2,3,4], Y. SUH[1,2,3,4];
[2]Dept. of Biomed. Sci., [3]Neurosci. Res. Inst., [4]Transplantation Res. Inst., [1]Seoul Natl. Univ. Col. of Med., Seoul, Korea, Republic of

Abstract: Local translation is a rapid process of synthesizing proteins from pre-existing synaptic mRNAs, independent of the cell body, which serves as a mechanism for supplying the synaptic proteome. Local translation plays a crucial role in various aspects of neuronal function, including development, synaptic plasticity, and information processing. Ribonucleoprotein particles (RNPs), such as stress granules and p-bodies, along with their associated RNA-binding proteins (RBPs), are known to participate in the regulation of local translation. While many neuronal mRNAs are thought to be transported to synapses in a translationally repressed state with RNPs, the precise molecular mechanisms underlying the specific stimuli that regulate local translation remain unclear. We examined the signals that regulate local translation by tracking puromycin-labeled nascent peptides in rat primary cortical neurons exposed to different neuronal stimuli. Our results revealed that treatment with glutamate, KCl, and forskolin induced alterations in both global and synaptic translation rates. In addition, specific neuronal RBPs, including G3BP1, Dcp1a, TDP-43, and FMRP, showed different translating ribosome-enriched sedimentation properties depending on the type of stimuli. These findings provide insight into the role of RNPs in the regulation of neuronal local protein homeostasis in response to activity.

Disclosures: D. Park: None. **Y. Suh:** None.

Poster

PSTR126. Cortical Network Dynamics

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR126.01/C53

Topic: B.07. Network Interactions

Support: University of Michigan Neuroscience Fellows Fund
University of Michigan Taubman Scholars Fund
Pritzker Neuropsychiatric Research Consortium NextGen Fund
Magnificent Michigan Neuroscience Fellowship

Title: Alertness and Learning Interact to Produce Brief Periods of Optimal Cognition in Mice

Authors: *A. SARAVANAN[1], A. GHIMIRE[1], M. DING[2], L. S. BUENO-JUNIOR[1], B. O. WATSON[1];
[1]Dept. of Psychiatry, Univ. of Michigan Med. Sch., Ann Arbor, MI; [2]Brandeis Univ., Waltham, MA

Abstract: Sensory discrimination in mice depends on both task proficiency and arousal states, meaning that response accuracy may vary with motivation and/or attention levels throughout the several minutes of a task session, even in well-trained animals. It is possible, therefore, that the interaction between two timescales of animal behavior - i.e., multi-day learning and minute/hour-scale fluctuations in alertness - produce short-lived windows of optimal response accuracy. Here we develop methods to quantify these optimal performance windows based on multiple non-invasive readouts in water-deprived head-fixed mice, including whisker-based sensory accuracy,

reward consumption, wheel running, pupil dilation, eyelid opening, and facial movements throughout 60-min task sessions in 14 days of training. Based on these non-invasive measures, we identified a predictable sequence of arousal states both within and across daily task sessions, including: (1) erratic impulsive-like behavior in the initial 10-20 min of all sessions, possibly due to high arousal and high motivation; (2) quiet-wake periods of low to no responding in the final 10-20 min of all sessions, possibly due to low arousal and satiation/tiredness; and (3) above-chance response accuracy at late training, particularly during brief 10-20 min periods in the middle of the task sessions, possibly due to a combination of intermediate arousal and late-training ability. We also recorded single-neuron activity across somatosensory cortex layers in a subset of animals, and obtained preliminary observations that sensory-evoked neuronal responses become stronger at late training, particularly either during or after high accuracy performance. These observations suggest that alertness and learning interact in predictable manners, which could be examined in future studies on altered brain states or cognitive dysfunctions.

Disclosures: A. Saravanan: None. **A. Ghimire:** None. **M. Ding:** None. **L.S. Bueno-Junior:** None. **B.O. Watson:** None.

Poster

PSTR126. Cortical Network Dynamics

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR126.02/C54

Topic: B.07. Network Interactions

Support: Naito Grant

Title: Optogenetic activation of dorsal raphe serotonin neurons induces a brain-wide activation of reward-related circuits

Authors: *H. T. HAMADA[1,2], Y. ABE[3], N. TAKATA[4], M. TAIRA[5], K. F. TANAKA[6], K. DOYA[7];
[1]Okinawa Inst. of Sci. & Technol., Onna, Okinawa, Japan; [2]Araya Inc., Tokyo, Japan; [3]Dept. of Neuropsychiatry, Keio Univ. Sch. of Med., Tokyo, Japan; [4]Neuropsychiatry, Keio University, Sch. of Med., Tokyo, Japan; [5]Dept. of Psychology, Univ. of California, Los Angles, Los Angeles, CA; [6]Dept. of Neuropsychiatry, Sch. of Medicine, Keio Univ., Tokyo, Japan; [7]Neural Computation Unit, Okinawa Inst. of Sci. and Technol., Onna Village, Japan

Abstract: Serotonin is a neuromodulator affecting many cognitive and behavioral functions and a key target for psychiatric treatments. To understand how the serotonin system regulates these functions through its extensive projections with diverse receptors, we stimulated dorsal raphe (DR) serotonin neurons in Tph2-ChR2 transgenic mice and observed the whole-brain BOLD responses by functional MRI with a 11.7T scanner with high S/N cryo coils. Following an acclimation protocol, optogenetic stimulation of DR serotonin neurons in awake mice resulted in a wide-spread activation including reward-related brain areas, including the orbitofrontal cortex

(OFC), medial prefrontal cortex (mPFC), caudate putamen (CPu), and ventral tegmental area (VTA) ($p<0.05$, multiple correction). A subsequent session confirmed consistent amplitudes and peak timing of BOLD responses in these areas (amplitude: $r=0.886$, $p<1e-8$; timing: $r=0.679$, $p<0.005$). Applying the same optogenetic stimulation under general anesthesia by isoflurane, we detected negative BOLD responses across the brain, including OFC, mPFC, retrosplenial cortex, CPu, and the hippocampal complex ($p<0.05$, multiple correction), consistent with a previous study that reported broad cortical BOLD signal deactivation under anesthesia (Grandjean et al. 2019). These results imply that general anesthesia reverses BOLD responses by DR serotonin activation from activation to inhibition. We further analyzed the BOLD responses using a multiple linear regression model incorporating the DR serotonin projection density and gene expression profiles of serotonin receptor subtypes 5HT1A, 5HT1B, 5HT1F, 5HT2A, and 5HT2C. Awake-state BOLD responses could be approximated by a model with negative weights for inhibitory 5HT1 receptors and positive weights for excitatory 5HT2 receptors ($p<0.05$, $R^2=0.66$). The BOLD response under general anesthesia correlated with an increased weight for the inhibitory 5HT1F receptor and a decreased weight for the excitatory 5HT2c receptor. Our study provides a novel functional anatomical perspective that DR serotonin elicits brain-wide activation of reward-related circuits, which is consistent with recent findings that DR serotonin neurons are activated during reward-oriented behaviors.

Disclosures: **H.T. Hamada:** None. **Y. Abe:** None. **N. Takata:** None. **M. Taira:** None. **K.F. Tanaka:** None. **K. Doya:** None.

Poster

PSTR126. Cortical Network Dynamics

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR126.03/C55

Topic: B.07. Network Interactions

Support: Department of Anesthesiology, University of Michigan Medical School, Ann Arbor, MI

Title: Effect of intravenous salvinorin A, the primary psychoactive molecule in Salvia divinorum, on brain network dynamics in rat

Authors: *E. R. HUELS[1,2,3], L. WANG[1], T. LIU[1], G. A. MASHOUR[1,2,3], D. PAL[1,2,3,4]; [1]Dept. of Anesthesiol., [2]Ctr. for Consciousness Sci., [3]Neurosci. Grad. Program, [4]Dept. of Mol. & Integrative Physiol., Univ. of Michigan, Ann Arbor, MI

Abstract: Salvinorin A, a kappa-opioid agonist and the primary psychoactive molecule in *Salvia divinorum*, has received little attention amid the recent psychedelic renaissance, which could be due to the virtual lack of preclinical brain dynamics data from animal studies to inform translational or clinical studies. Therefore, we determined the effect of intravenous administration of salvinorin A (2mg/kg) on brain dynamics in adult Sprague Dawley rats (n= 2

male, 1 female) using nondirectional (weighted phase lag index-wPLI) and directional (normalized symbolic transfer entropy-NSTE) measures of functional brain connectivity. The rats were instrumented to record high-density (30 channel) electroencephalogram (EEG) across the cortex and receive an intravenous bolus (5 minutes) infusion of salvinorin A or the vehicle control (14:26:60 DMSO:PEG400:saline). After 7-10 days of postsurgical recovery, EEG data were collected (0-300 Hz, sampling rate at 1 kHz) before, during, and after salvinorin A/vehicle infusion. EEG analyses were performed on artifact-free 1-5 minute EEG epochs before and immediately after salvinorin A or vehicle administration. The data are presented as percent change (mean ± SD) from the pre-salvinorin A/vehicle epoch. The most salient effects of salvinorin A occurred in the medium (65-125Hz) and high (125-155Hz) gamma bands. Salvinorin A increased wPLI in the medium gamma (24.55% ± 17.84%) and high gamma (61.11% ± 25.54%) ranges while the vehicle showed relatively small changes (medium gamma: 4.06% ± 15.64%; high gamma: -1.58% ± 4.25%). Salvinorin A also altered feedforward (parietal-to-frontal) and feedback (frontal-to-parietal) connectivity (NSTE), with decreases occurring in the medium gamma band (feedforward: -27.76 ± 4.96%; feedback: -25.43% ± 4.93) and increases in the high gamma band (feedforward: 50.62% ± 15.52%; feedback: 54.45% ± 17.49%). Vehicle administration did not produce similar changes in medium gamma (feedforward: -2.31% ± 2.61%; feedback: 4.24% ± 8.54%) or high gamma (feedforward: -3.4% ± 2.97%; feedback: -2.4% ± 6.12%) directed connectivity. Salvinorin A also led to decreased theta directed connectivity (feedforward: -39.67% ± 5.55%; feedback: -46.5% ± 7.45%), with much smaller decreases occurring after vehicle infusion (feedforward: -14.64% ± 26.42%; feedback: -26.54% ± 11.62%). To our knowledge, this is the first study to characterize the effects of salvinorin A on brain network dynamics in a rat model and will help inform future mechanistic and translational studies.

Disclosures: **E.R. Huels:** None. **L. Wang:** None. **T. Liu:** None. **G.A. Mashour:** None. **D. Pal:** None.

Poster

PSTR126. Cortical Network Dynamics

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR126.04/C56

Topic: B.07. Network Interactions

Support: Department of Anesthesiology, University of Michigan Medical School, Ann Arbor, MI

Title: Intravenous administration of 2,5 Dimethoxy-4-iodoamphetamine, a serotonergic psychedelic, produces active emergence from propofol anesthesia and restoration of spectral power and functional connectivity

Authors: C. W. FIELDS[1,2], E. R. HUELS[1,2,3], T. LIU[1], G. A. MASHOUR[1,2,3], *D. PAL[1,2,3,4]; [1]Dept. of Anesthesiol., [2]Ctr. for Consciousness Sci., [3]Neurosci. Grad. Program, [4]Dept. of Mol. & Integrative Physiol., Univ. of Michigan, Ann Arbor, MI

Abstract: Serotonergic psychedelics have been shown to produce an acute increase in wakefulness and decrease in sleep. At a systems level, serotonergic psychedelics enhance neurophysiological complexity and expand brain state repertoire, which is the opposite of that observed during anesthetic-induced unconsciousness. Based on this, we tested the hypothesis that a serotonergic psychedelic could induce active emergence from anesthesia. We assessed the effects of 2,5 Dimethoxy-4-iodoamphetamine (DOI), a 5-HT2A/2C agonist and psychedelic, on emergence from propofol anesthesia, spectral power, and functional connectivity. Adult Sprague Dawley rats (n=4 male) were equipped for high-density EEG recordings (30 channel) and intravenous delivery of propofol and DOI. After 7-10 days of postsurgical recovery, EEG was recorded during baseline wakefulness for 30 minutes, after which anesthesia was induced using intravenous propofol (1000 ug/kg/min) for 20 minutes. The propofol infusion rate was reduced after 20 minutes to 600 ug/kg/min, which was maintained for an additional 20 minutes. DOI (0.5mg/kg) was delivered intravenously over a 1-minute period during the continuous propofol infusion and rats were monitored for changes in behavior. EEG epochs of 1-5 minutes were selected from immediately before and after infusion of DOI during anesthesia to quantify the changes in absolute spectral power, magnitude-squared coherence, and directed functional connectivity (normalized symbolic transfer entropy) in the medium (65-125Hz) and high gamma (125-155Hz) bandwidth. The DOI infusion during continuous propofol anesthesia induced pronounced behavioral arousal leading to return of righting reflex (a surrogate for recovery of consciousness) in all four rats. EEG analyses (% change from pre-DOI anesthesia epoch) revealed increases in spectral power (medium gamma: 204.95% ± 139.87%, high gamma: 202.24% ± 119.31%) and coherence (medium gamma: 118.94% ± 14.6%, high gamma: 96.51% ± 15.43%). Similar increases were observed in normalized symbolic transfer entropy in both the feedforward (medium gamma: 350.18% ± 111.57%, high gamma: 197.31% ± 42.05%) and feedback (medium gamma: 428.13% ± 89.86%, high gamma: 193.99% ± 36.73%) direction. In a separate group of Sprague Dawley rats (n=4 male), the 5-HT2A antagonist volinanserin (0.025mg/kg intravenous bolus) was administered five minutes prior to DOI delivery and was shown to attenuate DOI-induced behavioral arousal and changes in EEG. These results provide preliminary evidence for psychedelic reversal of anesthesia.

Disclosures: C.W. Fields: None. **E.R. Huels:** None. **T. Liu:** None. **G.A. Mashour:** None. **D. Pal:** None.

Poster

PSTR126. Cortical Network Dynamics

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR126.05/C57

Topic: B.07. Network Interactions

Support: Tryp Therapeutics
Department of Anesthesiology, University of Michigan

Title: Intravenous psilocybin disrupts theta-gamma coupling and enhances frontoparietal networks in rat

Authors: *B. SILVERSTEIN[1,2,3], N. KOLBMAN[4,2,3], T. LIU[5], P. GUZZO[7], J. GILLIGAN[7], G. A. MASHOUR[5,2,3], G. VANINI[5,2,3], D. PAL[5,2,3,6];
[1]Univ. of Michigan, Ann Arbor, Detroit, MI; [2]Ctr. for Consciousness Sci., [3]Michigan Psychedelic Ctr., [4]Pharmacol., [5]Anesthesiol., [6]Dept. of Mol. & Integrative Physiol., Univ. of Michigan, Ann Arbor, MI; [7]Tryp Therapeut., Kelowna, BC, Canada

Abstract: Psilocybin is a serotonergic psychedelic that is being explored for therapeutic potential to treat mental health disorders, including depression and addiction. Large-scale alterations in frontoparietal networks have been observed in human fMRI and MEG studies of psilocybin. However, there are limited data on the effect of psychedelics, including psilocybin, on network dynamics in rodent brain, which is the preferred model for causal and mechanistic studies. Here, we used HD EEG (27 electrodes) recordings from rat cortex to determine dose-dependent effects of psilocybin on theta (4-10 Hz), medium gamma (65-95 Hz), and high gamma (95-150 Hz) brain network dynamics. Each rat (adult Sprague Dawley, n=6 male and 6 female) received three doses of psilocybin (0.1 mg/kg, 1 mg/kg, and 10 mg/kg) and saline as continuous intravenous infusion over 60 minutes, separated by at least one week and in a randomized order. We employed weighted phase-lag index (wPLI) based connectivity and node degree to assess the evolution of network organization within each band during the infusion. Phase-amplitude coupling (PAC) dynamics are associated with cognitive functions, such as memory and attention, and are altered following changes in arousal. We employed PAC to assess how psilocybin alters the time-dependency between theta and gamma activity. Linear mixed models were employed for global dynamics and follow-up FDR-corrected *t*-tests for channel-level changes. Psilocybin administration at 1 mg/kg and 10 mg/kg disrupted theta-medium gamma ($p<0.0001$) and theta-high gamma ($p<0.0001$) PAC at all electrodes across the cortex (FDR-corrected $p<0.05$); there was no statistical change in PAC after 0.1 mg/kg infusion ($p>0.05$). Global theta wPLI was increased in a dose-dependent fashion ($p<0.0001$) and was characterized by increased node degree in parietal channels (FDR-corrected $p<0.05$). Psilocybin did not change medium gamma global wPLI or node degree ($p>0.05$). Global high gamma wPLI increased during the administration of 1 mg/kg and 10 mg/kg doses ($p<0.0001$), characterized by increased frontal and parietal node degree (FDR-corrected $p<0.05$); there was no statistical change in frontoparietal node degree after 0.1 mg/kg infusion ($p>0.05$). Psilocybin infusion results in a dose-dependent reorganization of large-scale brain dynamics. The reorganization and increased frontoparietal network connectivity profiles parallel network changes observed in human fMRI studies of psilocybin, providing initial validation of the rodent model.

Disclosures: B. Silverstein: None. **N. Kolbman:** None. **T. Liu:** None. **P. Guzzo:** A. Employment/Salary (full or part-time); Tryp Therapeutics. **J. Gilligan:** A. Employment/Salary (full or part-time); Tryp Therapeutics. **G.A. Mashour:** None. **G. Vanini:** None. **D. Pal:** None.

Poster

PSTR126. Cortical Network Dynamics

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR126.06/Web Only

Topic: B.07. Network Interactions

Title: Eigenspace of Sparse Graph of Scalp EEG Explain Why a Fewer Electrodes Can Decode Better Than All!

Authors: *R. DEV, S. KUMAR, T. K. GANDHI;
Indian Inst. of Technol. Delhi, New Delhi, India

Abstract: Existing literature supports the idea that a smaller number of electrodes instead of all perform better at decoding the stimuli because of the exclusion of redundant and noisy electrodes, in EEG modality. However, it is not explainable why removing redundant electrodes can improve decodability. Also, the argument of 'noisy' channels, does not explain the improved accuracy since having noisy data does not imply poor accuracy. Most of these decoding algorithms use learning theory-based classifiers, hence, improvement in accuracy can be merely because of classifiers getting trained better, rather than because of electrodes being redundant or noisy. Therefore, we used an analytical model to understand what intrinsically causes the improved accuracy.
Initially, we construct a sparse graph from the running window of EEG signals and analyze its eigenspace. Then, we extract the sub-band characteristic vector (sCRV) time series, representing the graph's intrinsic structural dynamics. Distance between sCRV time-series of different stimuli is then observed to see how far they sit in graph eigenspace. To analyze how it varies with the number of electrodes, we plotted these time series for four sets of channels viz. 64, 32, 16, and 8. The more the distance between the sCRV of the condition, more is the decodability and hence better accuracy of decoding the stimulus. We used PhysioNet MI public dataset. We plotted the time series of the distance between the sCRV of consecutive time instants for both conditions alongside the time series of the distance between the sCRV of different MI conditions to see how far both conditions sit in eigenspace, averaging over 105 subjects with 3 trials for each. It can be seen from Figure 1, that reducing the number of channels, increases the distance between the two MI conditions. Henceforth the decodability improves with decreasing number of channels. Preliminarily, it can be said that improvement in accuracy might be because of the reduction in noise which is in line with the claims in the literature however further investigation is needed to conclusively find the cause.

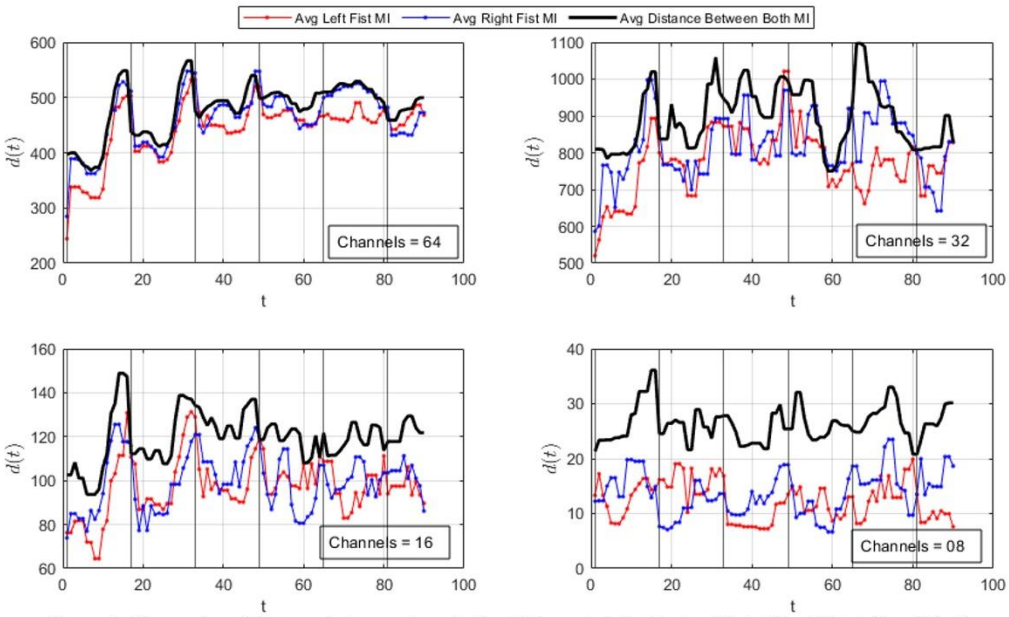


Figure 1: Timeseries of distance between the sub-Band Characteristic Vector 'Right Hand Fist MI' and 'Left Hand Fist MI' for various set of channels.

Disclosures: **R. Dev:** None. **S. Kumar:** None. **T.K. Gandhi:** None.

Poster

PSTR126. Cortical Network Dynamics

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR126.07/C58

Topic: B.07. Network Interactions

Support: GR 2024/11 -1

Title: Beyond oscillations? The potential of a diversified characterization of brain states changes

Authors: ***E. BALESTRIERI**[1], N. CHALAS[1], C. STIER[1], J. FEHRING[1], C. GIL ÁVILA[2], M. PLONER[3], J. GROSS[1];
[1]Univ. of Muenster, Muenster, Germany; [3]TU Muenchen, [2]TU Muenchen, Munich, Germany

Abstract: Our moment-to-moment conscious experience is paced by transitions between states, each one corresponding to a change in the electromagnetic brain activity. One consolidated analytical choice is to characterize these changes in the frequency domain, such that the transition from one state to the other corresponds to a difference in the strength of oscillatory power, often in pre-defined, theory-driven frequency bands of interest. Today, the huge leap in available computational power allows us to explore new ways to characterize electromagnetic brain activity and its changes. Here we leveraged an innovative set of features from the comprehensive highly comparative time-series analysis toolbox (hctsa, Fulcher and Jones, 2017).

On an MEG dataset with 29 human participants, we tested how these features described some of those state transitions known to elicit prominent changes in the frequency spectrum, such as eyes-closed vs eyes-open or the occurrence of visual stimulation. We then compared the informativeness of multiple sets of features by submitting them to a multivariate classifier (SVM). We found that the new features outperformed traditional ones in generalizing states classification across participants. Moreover, some of these new features yielded systematically better decoding accuracy than the power in canonical frequency bands that has been often considered a landmark in defining these state changes. Critically, we replicated these findings, after pre-registration, in an independent EEG dataset (N=210). In conclusion, the present work highlights the importance of a full characterization of the state changes in the electromagnetic brain activity, which takes into account also other dimensions of the signal on top of its description in theory-driven frequency bands of interest.

Disclosures: E. Balestrieri: None. **N. Chalas:** None. **C. Stier:** None. **J. Fehring:** None. **C. Gil Ávila:** None. **M. Ploner:** None. **J. Gross:** None.

Poster

PSTR126. Cortical Network Dynamics

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR126.08/C59

Topic: B.07. Network Interactions

Support: ERC StG 335880
ERC CoG 864491

Title: The causal structure of band-limited cortical dynamics

Authors: *P. HEGE[1,2,3,4,5], M. SIEGEL[2,3,4];
[1]Neural Dynamics and MEG, Univ. of Tuebingen, Tübingen, Germany; [2]Dept. Neural Dynamics and MEG, Hertie Inst. for Clin. Brain Research, Univ. of Tübingen, Tübingen, Germany; [3]Ctr. for Integrative Neuroscience, Univ. of Tübingen, Tübingen, Germany; [4]MEG Center, Univ. of Tübingen, Tübingen, Germany; [5]Mathematical Institute, Univ. of Tübingen, Tübingen, Germany

Abstract: The large-scale correlation of cortical activity may reflect interactions between brain regions. But instantaneous correlations are not informative about the dynamical and causal nature of such interactions. To address this, we investigated the dynamics of human cortical activity using source-reconstructed magnetoencephalography (MEG). We employed a novel orthogonalization approach to exclude volume conduction effects and systematically analyzed the time-lagged cross-correlation of neural activity across different frequency bands and the full cortical space. We focused on the time-asymmetric part of the cross-correlation to study causal effects. We found ubiquitous spatially and spectrally specific time-asymmetric correlations of cortical activity across the human brain. We devised a new constrained version of independent component analysis to identify components of these cross-correlations that can be interpreted as

causal patterns of lagged activation across cortical regions and frequency bands. We identified several fundamental causal patterns that were consistent across subjects, corresponded to spectrally specific interactions in well delimited cortical networks and were selectively modulated when subjects performed a cognitive task. Our results provide new insights into the causal structure of large-scale human brain dynamics, and open a new window to study these dynamics in the healthy and diseased brain.

Disclosures: P. Hege: None. **M. Siegel:** None.

Poster

PSTR126. Cortical Network Dynamics

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR126.09/C60

Topic: B.07. Network Interactions

Support: DFG EXC 307
DFG SI 1332/3
ERC StG 335880
ERC CoG 864491

Title: Region-specific relation between spiking and LFP dynamics in the primate brain

Authors: *A. IBARRA CHAOUL[1,2,3,4], C. VON NICOLAI[2,3,4], M. SIEGEL[2,3,4];
[1]Univ. of Tuebingen, Tuebingen, Germany; [2]Dept. Neural Dynamics and MEG, Hertie Inst. for Clin. Brain Res., University of Tübingen, Germany; [3]Ctr. for Integrative Neurosci., University of Tübingen, Germany; [4]MEG Ctr., University of Tübingen, Germany

Abstract: The local field potential (LFP) is a robust and widely used population measure of neural activity. However, basic questions concerning the LFP remain unclear. Are the temporal dynamics of the LFP, as reflected in its spectral power, specific for different brain regions? If so, does this specificity reflect the specific dynamics of spiking activity or of the relationship between spiking and the LFP? To address these questions, we performed large-scale simultaneous recordings of spiking activity and LFPs across several cortical and subcortical brain regions in macaque monkeys. Based on Wiener-Kolmogorov filtering, we decomposed the LFP power spectrum into the spectral information from the spike autocorrelation and from a kernel representing the relation between spiking and the LFP. We found that LFP dynamics are region specific, and that the kernels are more distinct between areas than spike autocorrelation functions. By further decomposing the kernel into its periodic and aperiodic components we found that the periodic, i.e., oscillatory part of the kernel captures most region-specific characteristics. Together, our results suggest that region-specific LFP dynamics are predominantly reflecting region-specific oscillatory spike-LFP coupling.

Disclosures: A. Ibarra Chaoul: None. **C. von Nicolai:** None. **M. Siegel:** None.

Poster

PSTR126. Cortical Network Dynamics

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR126.10/C61

Topic: B.07. Network Interactions

Support: NIH intramural program

Title: Spiking activity in cortical modules

Authors: *J. I. CHAPETON[1], K. SWIFT[1], S. INATI[1], K. A. ZAGHLOUL[2];
[1]NIH, Bethesda, MD; [2]NINDS, Bethesda, MD

Abstract: The synaptic connectivity of local cortical circuits places constraints on neural activity, and these constraints have consequences on how information can be stored and transmitted in cortical networks. One pattern of activity that has been observed in humans is the bursting of a population of neurons in a temporal sequence. These sequences appear during memory encoding, are reinstated during memory retrieval, and represent category specific information. Such a sequence-based code offers substantial combinatorial power for representing information, however, learning the set of all possible sequences in a large neural network is likely to be computationally intractable. One hypothesis is that long spike sequences are made up of subsequences much like how words are made up of syllables.

In a recent study we showed that local networks in the temporal lobe can be segregated into modules based on functional connectivity between LFP signals. By analyzing spiking responses during a categorization task, we also showed that neurons within a module share more information about sematic category compared to neurons across modules. The geometry of these modules, spatially contiguous ellipsoids about 1mm in diameter, is consistent with the size of a cortical column, raising the possibility that the observed spiking sequences are composed of subsequences that emerge from the activation of individual cortical columns. To test for this, we are analyzing a large dataset of local field potential recordings and single unit spiking activity captured from microelectrode arrays implanted in the human temporal lobe.

Specifically, we use the LFPs to determine the modular structure for each MEA and then assign each spike sorted neuron into a module based on the electrode it was recorded from. First, we find that while some population bursts can engage the entire MEA, others are constrained to neurons belonging to a single module. To quantify this, we calculated the neuronal synchrony between the neurons within module and find that it is significantly higher than the synchrony between random groups of neurons. We then treated the individual module bursts as binary signals and computed the cross-correlation between module bursts. We find that some pairs of modules have strong correlations in their bursting with lags of a few milliseconds. Together these preliminary results suggest that individual module bursts may reflect the activation of a cortical column, and that the array wide bursts may arise from the co-activation of several modules.

Disclosures: **J.I. Chapeton:** None. **K. Swift:** None. **S. Inati:** None. **K.A. Zaghloul:** None.

Poster

PSTR126. Cortical Network Dynamics

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR126.11/C62

Topic: B.07. Network Interactions

Support: Tiny Blue Dot Foundation

Title: Responses to cortical stimulation reveal thalamocortical state-dependent features in mice and humans

Authors: ***S. RUSSO**[1,2,3], L. D. CLAAR[3], L. MARKS[3], G. FURREGONI[1], F. ZAULI[1,4], G. HASSAN[1], M. SOLBIATI[1,4], S. SARASSO[1], M. ROSANOVA[1], I. SARTORI[4], A. PIGORINI[1], C. KOCH[3], M. MASSIMINI[1], I. REMBADO[3];
[1]Univ. of Milan, Milano, Italy; [2]Georgia Inst. of Technol., Atlanta, GA; [3]Allen Inst., Allen Inst., Seattle, WA; [4]Ctr. of Epilepsy Surgery "C. Munari", Niguarda Hosp., Milan, Italy

Abstract: Brief, direct cortical stimulation has been widely used in humans to treat neurological disorders (Sprengers, 2017; Little, 2013), restore lost neuronal functions (Gupta, 2023), and diagnose disorders of consciousness (Casarotto, 2016). Despite its broad use, we still do not know the circuits and mechanisms engaged by such transient brain stimulation, in particular the involvement of cortico-thalamic circuits (Van der Werf, 2006).
To fill this gap, we developed a unique experimental setup in head-fixed mice that combines a cortex-wide electroencephalography (EEG) array, with linear Neuropixels silicon probes (Jun, 2017) to simultaneously measure EEG and corticothalamic spiking activity evoked by direct cortical stimulation (Claar, 2023), together with optogenetic circuit manipulations.
Electrical stimulation (ES) of deep layers in the secondary motor cortex in awake mice induces a thalamocortical off-period followed by a thalamic rebound that in turn projects back to the cortex, generating a late EEG component at about 200 ms after stimulation onset. This thalamocortical loop is modulated by locomotion, such that when the animal is running the stimulation evokes thalamic bursts at lower frequency and less synchronous spikes, which correlates with a decrease in magnitude of the late evoked EEG component. Moreover, by combining electrical stimulation with timed optogenetic inhibition of the thalamus, we confirmed the causal relationship between thalamic rebound and late EEG component.
Follow-up experiments in humans revealed that sensorimotor tasks modulate the magnitude of the same late EEG component evoked by perturbation of the premotor area with ES in epileptic patients undergoing intracerebral evaluation and with transcranial magnetic stimulation (TMS) in healthy subjects. Despite the differences between species (humans-mice) and method of stimulation (ES-TMS), these results point towards a crucial contribution of the thalamo-cortical loop to the late response evoked by stimulation of premotor areas.
This study leverages on two noteworthy features: first, the use of a unique experimental setup in

mice that merges EEG signals - widely used in clinical settings - with high-density silicon probes; second, the development of experimental protocols translated from humans to mice and back. This suggests that the engagement of cortico-thalamo-cortical loops is preserved across species and stimulation methods. In future, this approach will lead to a better understanding of the neuronal circuits engaged by brain stimulation, potentially shining light on the fundamental mechanisms underlying its applications.

Disclosures: **S. Russo:** A. Employment/Salary (full or part-time); Manava Plus. **L.D. Claar:** None. **L. Marks:** None. **G. Furregoni:** None. **F. Zauli:** None. **G. Hassan:** None. **M. Solbiati:** None. **S. Sarasso:** None. **M. Rosanova:** None. **I. Sartori:** None. **A. Pigorini:** None. **C. Koch:** E. Ownership Interest (stock, stock options, royalty, receipt of intellectual property rights/patent holder, excluding diversified mutual funds); Intrinsic Powers Inc. **M. Massimini:** E. Ownership Interest (stock, stock options, royalty, receipt of intellectual property rights/patent holder, excluding diversified mutual funds); Intrinsic Powers Inc.. **I. Rembado:** None.

Poster

PSTR126. Cortical Network Dynamics

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR126.12/C63

Topic: B.07. Network Interactions

Support: NIH/MH127085
NIH/NINDS 113245
Tan-Yang Center for Autism Research at MIT
Stanley gift funding at the Broad Institute

Title: Distinct structural and functional connectivity of genetically segregated thalamoreticular subnetworks

Authors: N. HARTLEY[1,2], A. KROL[2], S. CHOI[1], N. ROME[1], S. PASQUALONI[1], C. JONES[1], R. KAST[2], G. FENG[2,1], *Z. FU[1,2];
[1]Broad Inst. of MIT and Harvard, Cambridge, MA; [2]MIT, Cambridge, MA

Abstract: The thalamic reticular nucleus (TRN) plays essential roles in sensory processing, arousal, attention, and cognition. Elucidating TRN cellular and circuit elements that control discrete behaviors is a prerequisite for understanding how TRN dysfunction contributes to distinct pathophysiological aspects of disease conditions. We have developed Cre mouse lines targeting genetically segregated populations of TRN neurons expressing the Spp1 and Ecel1 genes that engage first order (FO) and higher order (HO) thalamic nuclei, respectively. We show that Spp1 and Ecel1 neurons have distinct physiological features defined by substantial differences in both passive and active membrane properties. These TRN subnetworks are further delineated by distinct biases in top-down cortical and bottom-up thalamic inputs with significant differences in brain-wide synaptic convergence. Furthermore, we demonstrate that dysfunction

of each cell population results in the production of distinct cortical electroencephalography (EEG) impairments common to neuropsychiatric disorders, providing a link between discrete sensory processing abnormalities and dysfunction of segregated TRN subnetworks.

Disclosures: **N. Hartley:** None. **A. Krol:** None. **S. Choi:** None. **N. Rome:** None. **S. Pasqualoni:** None. **C. Jones:** None. **R. Kast:** None. **G. Feng:** None. **Z. Fu:** None.

Poster

PSTR126. Cortical Network Dynamics

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR126.13/D1

Topic: B.07. Network Interactions

Support: Whitehall Research Grant
NIH R01 NS103993
CDMRP PRMRP Discovery Award PR201842
T32 Ruth L. Kirschstein NRSA, NIH
Klingenstein-Simons Fellowship Award in Neuroscience
NIH R01 DC020459, NIDCD
NIH R21 Early Career Research Award NIDCD
NARSAD Young Investigator Award, Brain & Behavior Research Foundation
Emerging Research Grant, Hearing Health Foundation

Title: In-vivo optogenetic and calcium imaging for mapping the relative strength of synaptic connections

Authors: *R. U. GOZ[1], N. A. SCHNEIDER[2], R. S. WILLIAMSON[2], B. M. HOOKS[1]; [1]Dept. of Neurobio., [2]Dept. of Otolaryngology, The Univ. of Pittsburgh, Pittsburgh, PA

Abstract: Changes in synaptic strength are hypothesized to be a major locus of plasticity in brain circuits during development and skill learning. But *in-vivo* quantitative methods to track changes in synaptic strength in a cell type specific manner are needed to test which connections actually change their relative strength during plasticity. Using optogenetic approaches originally tested in *ex-vivo* cortical slices permits cell type specific optical stimulation. *In-vivo* calcium imaging allows quantification of responses in defined postsynaptic cell types to address specificity of connections. Furthermore, imaging can be repeated over time, permitting tracking of plasticity following learning or degeneration. In these experiments, we quantify synaptic input to primary motor cortex (M1) pyramidal neurons and interneurons from two major excitatory inputs: primary somatosensory cortex (S1) and posterior thalamus (PO). We demonstrate that this *in-vivo* approach matches the layer- and cell type-specific results of monosynaptic input mapping in brain slices for parvalbumin-positive (PV+) interneurons and pyramidal neurons in cortical layers 2/3 and upper 5A. Specifically, that thalamic axonal stimulation is increasing

input on PV+ cells relative to pyramidal cells with cortical depth. While somatosensory axonal stimulation provides increasing input to pyramidal cells and less input to PV+ cells with increasing cortical depth. We monitor the stability of these connections over time. This approach provides specific information of brain behavior in a live, non-anesthetized animal and can be extended to map any cortical or subcortical inputs to genetically-identified cell populations, providing the ability to longitudinally track cortical plasticity in numerous contexts including learning and neurodegeneration.

Disclosures: R.U. Goz: None. **N.A. Schneider:** None. **R.S. Williamson:** None. **B.M. Hooks:** None.

Poster

PSTR126. Cortical Network Dynamics

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR126.14/D2

Topic: B.07. Network Interactions

Support: NIMH grant MH123748
NIMH grant MH122379
NIMH grant MH126548
Taylor Family Institute for Innovative Psychiatric Research
Bantly Foundation

Title: Periodic and aperiodic changes to cortical EEG in response to pharmacological manipulation

Authors: *S. SALVATORE[1], P. M. LAMBERT[1], A. BENZ[1], N. R. RENSING[2], M. WONG[2], C. F. ZORUMSKI[1], S. MENNERICK[1];
[1]Psychiatry, [2]Neurol., Washington Univ. in St.Louis, St.Louis, MO

Abstract: Cortical electroencephalograms (EEG) may help understanding of neuropsychiatric illness and new treatment mechanisms, given applicability of cortical EEG across species. The aperiodic component (1/*f*) of EEG power spectra is often treated as noise, but recent studies have highlighted its potential significance for brain function. Specifically, changes to the aperiodic exponent of power spectra may be associated with changes in excitation/inhibition (E/I) balance, a concept linked to antidepressant effects, epilepsy, autism, and other clinically important conditions. One confound of previous studies is behavioral state, because factors associated with behavioral state other than E/I balance may alter EEG parameters. Thus, to test the robustness of the aperiodic exponent as a predictor of E/I balance, we analyzed active exploration in mice using video EEG following various pharmacological manipulations with fitting oscillations & one over f (FOOOF) algorithm. We found that $GABA_A$ receptor ($GABA_AR$) positive allosteric modulators increased the aperiodic exponent, consistent with the hypothesis that an increased exponent signals enhanced cortical inhibition, but other drugs did not follow the prediction.

Ketamine produced no change in the exponent parameter at drug doses that robustly altered oscillatory components. $GABA_AR$ antagonists at sub-convulsive doses yielded counterintuitive behavioral sedation, increased the aperiodic exponent in the sedated state, and failed to decrease it upon return to active wake. To more conclusively tilt E/I balance toward excitation, we selectively inhibited parvalbumin (PV) interneurons with a designer receptor exclusively activated by designer drugs (DREADD). Contrary to our expectations and studies demonstrating increased cortical activity following PV inhibition, circuit disinhibition with a DREADD increased the aperiodic exponent. We conclude that the aperiodic exponent of EEG power spectra does not yield a reliable marker of E/I balance. Alternatively, the concept of E/I state may be sufficiently oversimplified that it cannot be mapped readily onto an EEG parameter.

Disclosures: S. Salvatore: None. **P.M. Lambert:** None. **A. Benz:** None. **N.R. Rensing:** None. **M. Wong:** None. **C.F. Zorumski:** E. Ownership Interest (stock, stock options, royalty, receipt of intellectual property rights/patent holder, excluding diversified mutual funds); Sage Therapeutics. F. Consulting Fees (e.g., advisory boards); Sage Therapeutics. **S. Mennerick:** None.

Poster

PSTR126. Cortical Network Dynamics

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR126.15/D3

Topic: B.07. Network Interactions

Support: NIH National Institute of General Medical Sciences grant R01GM134363-01

Title: Aperiodic electrophysiological activity tracks spiking statistics between behavioral states

Authors: *S. FIGUEROA[1,2,3], M. W. PRESTON[4], B. VOYTEK[4,5,3,6];
[2]Sch. of Biol. Sci., [3]Halıcıoğlu Data Sci. Inst., [4]Neurosciences Grad. Program, [5]Dept. of Cognitive Sci., [6]Kavli Inst. for Brain and Mind, [1]UCSD, La Jolla, CA

Abstract: Electrical brain activity can be measured at various temporal and spatial scales. For instance, mesoscopic signals such as electroencephalography (EEG) and local field potentials (LFP) reflect activity across populations of neurons. Notably oscillations in EEG and LFP have been associated with mechanisms of memory and cognition. However, emerging research has shown that non-oscillatory, aperiodic activity also serves as a biomarker of disease, age, cortical region, and cognitive state. Although the significance of aperiodic activity is strongly supported, the underlying mechanisms and physiological origin have not been fully characterized. In this study, we leveraged an open dataset (Allen Institute for Brain Science: Visual Coding - Neuropixels dataset) to investigate the relationship between population spiking activity and aperiodic LFP activity. Here we show that aperiodic LFP activity indexes the rate and synchrony of population spiking activity within and between cognitive and behavioral states. Our current

results support previous findings that broadband LFP power reflects the firing rate of a population. Specifically, we found a strong negative correlation between rate and low frequency power with a concomitant strong positive correlation at high frequency ranges, indicative of a singular, aperiodic process. Surprisingly, spike synchrony was found to be negatively correlated with the aperiodic exponent of the LFP, contrary to the predictions of previous models i.e. greater spike synchrony was associated with flatter power spectra. These findings support the idea that aperiodic EEG and LFP activity is a physiologically meaningful signal, providing information about the underlying population spiking statistics. Further investigation into the aperiodic components of electrical brain waves has the potential to provide valuable information surrounding cognition not present in oscillatory activity. More specifically, characterizing the physiological origin of aperiodic activity will advance our understanding of its functional role in cognition and disease.

Disclosures: S. Figueroa: None. **M.W. Preston:** None. **B. Voytek:** None.

Poster

PSTR126. Cortical Network Dynamics

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR126.16/D4

Topic: B.07. Network Interactions

Support: NSF1912352
NIHR15NS116742
CNPq 425037/2021-5

Title: Cortical state rate coding in the primary visual cortex

Authors: *A. FONTENELE[1], S. SOOTER[1], W. SHEW[1], N. A. VASCONCELOS[2];
[1]Univ. of Arkansas, Fayetteville, AR; [2]Biomed. Engin., Federal Univ. of Pernambuco, Recife, Brazil

Abstract: Nearly a century ago, Lord Adrian and colleagues, based on pioneering recordings of sensory and motor neurons, proposed that stimulus properties are encoded by spiking rates. Fueled by Hubel's new Tungsten, in the second half of the 1900s, Hubel and Wiesel demonstrated rate coding in isolated neurons in the primary visual cortex. Recently, with the simultaneous recordings of neuronal populations, a novel sensory coding perspective emerged, which expanded the understanding of the nervous system beyond what could be reached from recordings of isolated neurons. Based on Caton's recordings in vivo, back in the XIX century, we know that patterns of electrical activity in the brain surface (currently termed cerebral cortex) change as the time goes by. Classes of different patterns in the cortical activity define different cortical states with a corresponding dynamic diversity, mainly based on spiking activity. More recently, several studies have devoted attention to understanding how brain functions are dependent on those cortical states. Furthermore, using few discrete cortical states, contemporary

research has revealed the modulation of cortical state over individual cells located in the primary sensory cortex. In our study, we extend this prior work by examining the spiking rate modulation in single neurons in the primary visual cortex, along a more diverse set of cortical states, based on the level of spiking variability in their large neuronal populations. We found individual cells with a spiking rate tuned to specific cortical states. Based on unsupervised classification, we found a relatively small number of classes of spiking rate modulation by cortical states. Furthermore, those different classes of cells display significant statistical differences among spiking properties. Moreover, the average pairwise spiking correlations were stronger within classes than across classes, in both states: synchronous and asynchronous.

Disclosures: **A. Fontenele:** None. **S. Sooter:** None. **W. Shew:** None. **N.A. Vasconcelos:** None.

Poster

PSTR126. Cortical Network Dynamics

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR126.17/D5

Topic: B.07. Network Interactions

Support: NIH Grant R15NS116742
NSF Grant 1912352

Title: Criticality during awake state, supercriticality during deep sleep

Authors: ***S. SOOTER**[1], A. FONTENELE[2], N. A. VASCONCELOS[3], W. L. SHEW[2]; [1]Physics, Univ. of Arkansas Fayetteville, Fayetteville, AR; [2]Physics, Univ. of Arkansas, Fayetteville, AR; [3]Biomed. Engin., Champalimaud Ctr. for the Unknown, Recife, Brazil

Abstract: It has long been known that collective neural activity in cerebral cortex exhibits large synchronous fluctuations during slow-wave sleep. Studies of population activity in awake animals have also revealed diverse coordinated fluctuations, sometimes called neuronal avalanches. How do the slow waves during deep sleep differ from neuronal avalanches in the awake state? Which of these states, if any, is consistent with the criticality hypothesis, which predicts that avalanches are power-law distributed? Here, we analyzed extracellular electrophysiological recordings of unit activity from primary visual cortex of freely behaving mice, seeking signatures of criticality in both the NREM sleep and awake states. In the awake state, we found power-law avalanche size and duration distributions whose exponents obey the crackling noise scaling relation. In addition we found that the awake-state population activity exhibited a slowly decaying autocorrelation function, an approximately 1/f power spectrum, and a DFA exponent near unity. Together, these observations are strong evidence that awake mouse cortex is, indeed, close to criticality. In NREM sleep, on the other hand, we found that the avalanche size and duration distributions were poorly fit by power-laws. Moreover, the long-range temporal correlations observed in the awake state weren't present in NREM sleep, as we found that the population activity had a quickly decaying autocorrelation function, a nearly flat

power spectrum, and a DFA exponent close to that of a completely uncorrelated time series (0.5). Our observations suggest that the cortex shifts from criticality toward a supercritical state as the animal transitions from waking to NREM sleep.

Disclosures: S. Sooter: None. **A. Fontenele:** None. **N.A. Vasconcelos:** None. **W.L. Shew:** None.

Poster

PSTR126. Cortical Network Dynamics

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR126.18/D6

Topic: B.07. Network Interactions

Support: NSF Grant 1912352
NIH Grant R15NS116742

Title: Low dimensional criticality embedded in high dimensional awake brain dynamics

Authors: A. FONTENELE, S. SOOTER, K. NORMAN, S. GAUTAM, ***W. SHEW**;
Univ. of Arkansas, Fayetteville, AR

Abstract: Cerebral cortex has been hypothesized to operate close to criticality - at the tipping point between order and disorder. This hypothesis offers an explanation of the observed complexity of cortical dynamics and is important because of potential computational advantages near criticality. However, in the awake state, when cortex most needs computation, experimental evidence for criticality has been inconsistent, especially when considering high precision measurements, i.e. spikes of many single neurons measured with millisecond resolution. The inconsistency of previous reports casts doubt on the possibility that awake cortex operates near criticality. Here we show that discrepant previous reports of critical phenomena in the brain may be reconciled by considering dimensionality and dimensionality reduction of brain dynamics. Indeed, fundamental physics of critical phenomena emphasizes the importance of coarse-graining of observables, which is a type of dimensionality reduction. We show that coarse graining over neurons and time is a type of dimensionality reduction which reveals low-dimensional critical dynamics in a prominent subspace (first few principal components) of awake cortical dynamics.

Disclosures: A. Fontenele: None. **S. Sooter:** None. **K. Norman:** None. **S. Gautam:** None. **W. Shew:** None.

Poster

PSTR126. Cortical Network Dynamics

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR126.19/D7

Topic: B.07. Network Interactions

Support: NIH Grant R01MH112746
NIH Grant R01MH108590
SFARI Pilot Award

Title: Personalized circuit modeling captures leading modes of inter-individual variation in cortical functional connectivity

Authors: *R. A. COOPER[1], M. DEMIRTAS[2], J. B. BURT[3], A. HOWELL[3], J. JI[3], A. ANTICEVIC[4], J. D. MURRAY[5];
[1]Physics, [2]Psychiatry Dept., [4]Psychiatry, [5]Dept. of Psychiatry, [3]Yale Univ., New Haven, CT

Abstract: Substantial cortical variation exists across human beings, both anatomically and functionally. This variation is subject-specific, stable across time, and macroscopically observable via various modalities, including magnetic resonance imaging (MRI) and behavioral measures. However, the neural circuit mechanisms underlying across-individual variation remain unclear. An approach to bridging these mechanistic gaps is using biophysically-based neural circuit models of large-scale brain dynamics which can be quantitatively fit to empirical neuroimaging data. In this study we have utilized a circuit model with neurobiologically interpretable parameters to model functional connectivity (FC) at the individual level in healthy subjects. We evaluate the leading modes of variation in the empirical functional data, finding that our model is well-suited to capture variation both within and across subjects.
We selected a large cohort of subjects (N=842) from the Human Connectome Project, from which we generated parcellated, resting-state, empirical FC matrices. Using principal component analysis (PCA), we evaluate leading components of variation in two ways: across the 842 subjects and across time via the four functional scans for each subject. We find that the three leading principal components (PCs) of both within- and across-subject variation follow interpretable topographic patterns. Within-subject variation is strongly correlated with but distinct from across-subject variation. We applied our modeling framework to the individual-level functional MRI data as well. We explore patterns of sensitivity to parameter perturbation in the simulated FCs and evaluate model expressivity by geometrically quantifying how well these patterns of sensitivity capture the leading PCs across subjects.
This framework provides a principled approach to extending computational models and revealed a straightforward mapping between model parameters and and the leading modes of variation. In particular, this framework provides a rationale for a key component of this model: its assumption that local circuit properties are heterogeneous across the cortex, following a large-scale gradient related to cortical hierarchy. Further, we analyze other leading components of variation between and within individuals to incorporate corresponding structure into the model via a variety of physiological processes, which may help to indicate biological factors that strongly contribute to functional variation.

Disclosures: **R.A. Cooper:** None. **M. Demirtas:** None. **J.B. Burt:** None. **A. Howell:** None. **J. Ji:** None. **A. Anticevic:** None. **J.D. Murray:** None.

Poster

PSTR126. Cortical Network Dynamics

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR126.20/D8

Topic: B.07. Network Interactions

Support: Grants-in-aid for Scientific Research

Title: Analysis of the occurrence rate of neural activity patterns in a cultured neuronal networkusing instantaneous-spatial-pattern representation.

Authors: *H. ASADA[1], *H. ASADA[1], S. KUDOH[2];
[1]Sci. and Technol., Kwansei Gakuin Univ., Sanda-shi, Japan; [2]Sci. and Technol., Kwansei Gakuin Univ., Sanda, Hyougo, Japan

Abstract: The electrical activity evoked by inputs to a neuronal network is thought to represent information from the outside world. This activity forms a spatiotemporal pattern of the firing of synchronized neurons. In most previous studies, neuronal activity was analyzed by splitting the temporal sequence of neuronal activity into segments based on an empirically determined time window. However, this analysis time window and its phase were the same for all recorded activity trains (electrodes), which led to extracting neuronal activity patterns that varied with the time window width. Therefore, We hypothesized that information from the outside world is represented by a combination of serial signal propagation pathways in a neuronal network., and attempted to elucidate the co-occurrence of neuronal signal propagation pathways. We used a multi-point electrode array to measure action potential spike trains with high reproducibility from multiple channels. Then we sliced these spike trains into an instantaneous-time window of 1 ms width, which are shorter than the time required for synaptic transmission, and extracted a spatial pattern from each slice. We called these spatial patterns "instantaneous spatial pattern," which are footprints of the signal transmission by pecific neural pathways composed of synaptically connected neurons.. The instantaneous spatial patterns are independent of the width of the analysis time window, ensuring that the patterns do not involve direct functional synaptic connections between simultaneousl neural activities within each slice. Thus, the instantaneous-spatial-patterns represent time slices of the co-occurring neural signal propagation pathways. We stimulated the neuronal network and analyzed the instantaneous-spatial-patterns for spike trainsat multiple recording electrodes. The results showed that neuronal signals induced by stimulation propagate in parallel and that their co-occurrence patterns vary even for the same stimulus. The output pattern of the input signal is expected to change depending on the combination of co-activated neuronal signal propagation pathways.Therefore, we analyzed how the probability of firing patterns changed according to the relationship between upstream and downstream instantaneous spatial patterns.

Disclosures: **H. Asada:** None. **H. Asada:** None. **S. Kudoh:** None.

Poster

PSTR126. Cortical Network Dynamics

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR126.21/D9

Topic: B.07. Network Interactions

Title: Characterization of the electrophysiological properties of hallucinogenic and non-hallucinogenic compounds using large datasets.

Authors: *D. DVORAK[1], B. J. DEARNLEY[2,3], A. KRUEGEL[1], M. OKUN[2,3];
[1]Gilgamesh Pharmaceuticals Inc., New York, NY; [2]Univ. of Sheffield, Sheffield, United Kingdom; [3]Univ. of Nottingham, Nottingham, United Kingdom

Abstract: Serotonin 5-HT2A receptor agonists show promising therapeutic potential in the treatment of several psychiatric disorders, including depression, substance abuse, and post-traumatic stress disorder. However, the molecular, cellular, and circuit-level events downstream of 5-HT2A receptor activation that lead to the rapid and durable therapeutic effects of these compounds remain enigmatic. While the long-term therapeutic benefits might be associated with neuroplastic changes in cortical networks, the acute effects are likely important as well. The majority of prior electrophysiology studies aiming to describe the acute, 'drug-on' state have only involved the analysis of single compounds at limited doses and in a single brain area. This narrow approach limits the ability to generalize the findings of such studies across different 5-HT2A ligands or to apply them more broadly to explain therapeutic or other behavioral effects. To overcome the constraints inherent in previous work, we implemented the use of Neuropixels probes to record the acute dose-related effects of structurally diverse hallucinogenic and non-hallucinogenic 5-HT2A receptor agonists as well as 5-HT2A receptor antagonists from thousands of neurons across number of brain regions in awake head-fixed mice able to run in a 2D arena. These recordings allowed the assessment of changes in neural activity after a compound's administration using analysis of individual neuron firing rates, co-firing between pairs of cells, power changes in local field potentials, and spike-field coherence. We demonstrate that the drug category and its putative hallucinogenic properties can be identified using electrophysiological measures. This research represents a significant step towards understanding the extent to which the acute electrophysiological effects of diverse hallucinogenic and non-hallucinogenic compounds can be generalized. Such understanding, in turn, facilitates the rapid evaluation of novel compounds, enabling predictions about their potency, efficacy, and ability to induce hallucinogenic effects.

Disclosures: **D. Dvorak:** A. Employment/Salary (full or part-time); Gilgamesh Pharmaceuticals Inc.. E. Ownership Interest (stock, stock options, royalty, receipt of intellectual property rights/patent holder, excluding diversified mutual funds); Gilgamesh Pharmaceuticals Inc. **B.J. Dearnley:** B. Contracted Research/Research Grant (principal investigator for a drug study, collaborator or consultant and pending and current grants). If you are a PI for a drug study, report that research relationship even if those funds come to an institution.; Gilgamesh

Pharmaceuticals Inc. **A. Kruegel:** A. Employment/Salary (full or part-time); Gilgamesh Pharmaceuticals Inc.. E. Ownership Interest (stock, stock options, royalty, receipt of intellectual property rights/patent holder, excluding diversified mutual funds); Gilgamesh Pharmaceuticals Inc. **M. Okun:** B. Contracted Research/Research Grant (principal investigator for a drug study, collaborator or consultant and pending and current grants). If you are a PI for a drug study, report that research relationship even if those funds come to an institution.; Gilgamesh Pharmaceuticals Inc.

Poster

PSTR126. Cortical Network Dynamics

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR126.22/D10

Topic: B.07. Network Interactions

Support: BBSRC BB/P020607/1
BBSRC 2265967
Springboard award SBF002\1045

Title: Brain state transitions primarily impact the spontaneous rate of slow-firing neurons

Authors: B. J. DEARNLEY, M. SHAW, M. DERVINIS, *M. OKUN;
Univ. of Sheffield, Sheffield, United Kingdom

Abstract: The spontaneous firing rates of individual neurons are modulated by brain state. We examined how such modulation impacts the overall distribution of firing rates in neuronal populations of neocortical, hippocampal and thalamic areas in mice. We considered two naturally occurring categories of brain state transitions, namely sleep-wakefulness transitions and changes in arousal level in awake mice, along with two categories of pharmacologically induced brain state changes, namely transitions from a drug-free baseline state to a psychedelic state induced by 5-HT2A receptor agonist, and to a sub-anesthetic, dissociation-like state induced by NMDA antagonist. Our key findings can be summarized using the joint distribution across the neuronal population of rate modulation and mean log-rate, which captures the quantitative relationship between magnitude of firing rate change and baseline firing rate. For a neuron having firing rates r^A and r^B in brain states A and B, we defined its rate modulation index upon A-to-B transition as $\log(r^B) - \log(r^A)$, while its mean log-rate is $(\log(r^B) + \log(r^A))/2$. We used log-rates because firing rates are logarithmically scaled, with a bell-shaped log-rate distribution across the population. Across all the examined combinations of brain state transition categories and forebrain areas, we find that the joint distribution of rate modulation index and mean log-rate has a shape of a left-tilted ‘V’ (i.e., ‘>’), with high variability of modulation across slow-firing neurons and low variability across fast-firing neurons. (We verified that this is a genuine biological effect, not caused by the statistical bias of inherently noisier firing rate estimates for slow-firing neurons). As a result, the slow-firing neurons are on average more strongly modulated than the fast-firing ones. These differences in modulation variance and magnitude

along the firing rate spectrum also hold for upregulated and downregulated subpopulations considered separately. We further demonstrate that this modulation structure is linked to the left-skewed distribution of firing rates on the logarithmic scale and is recapitulated by bivariate log-gamma, but not Gaussian, distributions. Finally, the lack of interaction between modulations driven by two distinct categories of brain state transitions - arousal and a psychedelic drug in our case - indicates that the subpopulation of malleable slow-firing neurons can be unique to each category. Our findings indicate that a preconfigured log-rate distribution with rigid fast-firing neurons and a long left tail of malleable slow-firing neurons is a generic property of forebrain neuronal circuits.

Disclosures: B.J. Dearnley: None. **M. Shaw:** None. **M. Dervinis:** None. **M. Okun:** None.

Poster

PSTR126. Cortical Network Dynamics

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR126.23/D11

Topic: B.07. Network Interactions

Title: Circuit mechanisms underlying the control of prefrontal cortical activity by the basal amygdala

Authors: *D. MAGYAR[1,2,3], P. NAGY-PÁL[2,3], J. M. VERES[3], Z. REÉB[3,4], R. KARLÓCAI[1], N. HÁJOS[1,3];
[1]Indiana Univ., Bloomington, IN; [2]Semmelweis Univ., Budapest, Hungary; [3]ELRN Inst. of Exptl. Med., Budapest, Hungary; [4]ELTE Eötvös Loránd Univ., Budapest, Hungary

Abstract: Basal amygdala (BA) gives rise to one of it's major excitatory inputs into the dorsal part of the medial prefrontal cortex (dmPFC), which projects back heavily to the same amygdala nucleus. These projections play a key role in a variety of behavioral functions, including the regulation of memory-related and emotional processes and become dysfunctional in neuropsychiatric diseases. Since it is still not completely understood how amygdala output controls spiking activity within the dmPFC, in this study we aimed to uncover the underlying circuit mechanisms. To examine the circuit operation driven by amygdalar afferents, we first used in vivo multichannel electrophysiological recordings combined with optogenetics in awake, head-fixed mice. We observed that photoactivation of amygdalar inputs both excited and inhibited neuronal activity in dmPFC neurons in separate populations of cells, in addition to unaltering about half of the recorded units. Using anterograde viral tracing, we found that amygdala projections targeted both pyramidal cells and interneurons in the dmPFC. Whereas all types of pyramidal neurons defined by their projection areas were innervated by amygdala afferents, they targeted predominantly parvalbumin-expressing basket cells among GABAergic inhibitory cells. Interestingly, all pyramidal cells that received amygdala inputs were also targeted by amygdala-innervated parvalbumin-expressing basket cells. Using combined optogenetics and electrophysiology, we validated that excitation of amygdala-targeted

parvalbumin basket cells can reliably suppress spiking in the dmPFC. Using dual optogenetic approach, we confirmed that parvalbumin-expressing basket cells in the dmPFC were primarily responsible for suppressing neuronal activity upon amygdala afferent stimulation. Applying trains of afferent stimulation revealed that amygdala inputs could drive the firing of both interneurons with narrow spikes and pyramidal cells more reliable at 20Hz than 5Hz. Our results uncovered that amygdala-driven feedforward inhibition is accomplished via parvalbumin-expressing basket cells, which innervated amygdala-input receiving pyramidal cells, too. Thus, feedforward excitation and inhibition driven by amygdala afferents converge on the same dmPFC pyramidal cells. This structural motif may ensure a precise timing of dmPFC neuronal activity dictated by the basal amygdala.

Disclosures: D. Magyar: None. **P. Nagy-Pál:** None. **J.M. Veres:** None. **Z. Reéb:** None. **R. Karlócai:** None. **N. Hájos:** None.

Poster

PSTR126. Cortical Network Dynamics

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR126.24/D12

Topic: B.07. Network Interactions

Support: Wallenberg Fellowship from the Knut and Alice Wallenberg Foundation
ERC Starting grant
Swedish Brain Foundation (Hjärnfonden)
Swedish Medical Research Council (VR-M)
Singapore Ministry of Education [MOE2015-T2-2-095]
StratNeuro postdoctoral grant
Karolinska Institute doctoral grant.
Nanyang Technological University - Karolinska Institutet Joint PhD Programme

Title: Pre- and Postsynaptic Determinants of Functional Connectivity Between the Claustrum and Anterior Cingulate Cortex

Authors: *R. DE LA TORRE MARTINEZ[1], Z. CHIA[1,2,3], A. TOKARSKA[1], J. FROST-NYLEN[1], G. AUGUSTINE[2], G. SILBERBERG[1];
[1]Neurosci., Karolinska Inst., Stockholm, Sweden; [2]Lee Kong Chian Sch. of Med., [3]Sch. of Biol. Sci., Nanyang Technological Univ., Singapore, Singapore

Abstract: The claustrum (CLA), a brain nucleus positioned between the insula and lateral striatum, is implicated in diverse behaviors. The underlying mechanisms involve connectivity between the claustrum and cortical regions, including the anterior cingulate cortex (ACC). While CLA projection neurons are predominantly glutamatergic, previous studies have suggested an inhibitory impact of the claustrum on cortical targets. However, the specific synaptic connections

between different CLA and cortical cell types remain unknown. In this study, we employed in vivo and ex vivo electrophysiology and optogenetics to elucidate the functional organization of the CLA-ACC pathway based on pre- and postsynaptic populations. Optogenetic stimulation of CLA neurons in awake mice evoked multiphasic excitatory and inhibitory responses in ACC, contingent upon the stimulated CLA population, cortical layer and cell type. Paired recordings in ACC revealed monosynaptic responses in pyramidal cells and various interneurons following photostimulation of CLA-ACC synaptic terminals. Notably, CLA axons formed monosynaptic connections throughout all layers of ACC, but the synaptic response probability and strength varied depending on the type of CLA projection, target layer in ACC, and postsynaptic neuron type. This intricate organization of the CLA-ACC pathway sheds light on the complex influence of CLA on ACC and other cortical regions, bridging gaps in the field and enhancing our understanding of the functional role played by the claustrum in cortical function.

Disclosures: R. de la Torre Martinez: None. **Z. Chia:** None. **A. Tokarska:** None. **J. Frost-Nylen:** None. **G. Augustine:** None. **G. Silberberg:** None.

Poster

PSTR126. Cortical Network Dynamics

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR126.25/D13

Topic: B.07. Network Interactions

Title: Behavioural state-dependent modulation of claustrocortical communication

Authors: *A. D. DO[1], B. MARRIOTT[2], J. C. JACKSON[1,2];
[1]Physiol., [2]Neurosci. and Mental Hlth. Inst., Univ. of Alberta, Edmonton, AB, Canada

Abstract: The claustrum is a small subcortical brain region densely and reciprocally connected with the entire cortex. Previous optogenetic studies revealed that the claustrum provides feedforward inhibition to the cortex by directly activating cortical interneurons. However, the significance of this inhibition is unclear, partially because the behavioural contexts recruiting this inhibition have not been defined. We performed *in vivo* two-photon calcium imaging of claustrum neurons projecting to the retrosplenial cortex (CLA_{RSC}) in head-fixed mice behaving on a treadmill. We found that CLA_{RSC} neurons are mostly active during quiet wakefulness and less active or even silent during active wakefulness. Then, we optogenetically activated CLA_{RSC} neurons using the excitatory opsin ChR2 in mice spontaneously transitioning between rest and locomotion. While the optogenetic activation of CLA_{RSC} neurons produced a robust negative field potential in the retrosplenial cortex during rest, we observed an attenuation of the amplitude of this negative potential when claustrum activation occurred during periods of locomotion. Together, our results suggest that the relay of information from the claustrum to the retrosplenial cortex declines during active wakefulness compared to quiet wakefulness via at least two mechanisms: 1) A physiological decrease in the activity of these cells and 2) A reduction in the communication efficacy from the claustrum to the retrosplenial cortex.

Disclosures: **A.D. Do:** None. **B. Marriott:** None. **J.C. Jackson:** None.

Poster

PSTR126. Cortical Network Dynamics

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR126.26/D14

Topic: B.07. Network Interactions

Support: CIHR Grant: 426485
NSERC: RGPIN-2020-05988
Brain Canada Foundation
The Scottish Charitable Rite Foundation

Title: Bidirectional chemogenetic modulation of claustral activity causes altered cortical dynamics in anesthetized mice

Authors: *R. ZAHACY[1], Y. MA[2], I. R. WINSHIP[2], J. JACKSON[3], A. W. CHAN[2];
[1]Neurosci. and Mental Hlth. Inst., [2]Psychiatry, [3]Physiol., Univ. of Alberta, Edmonton, AB, Canada

Abstract: The claustrum is a thin, long, subcortical structure, located bilaterally with dense connections to the cortex. Here we bi-directionally chemogenetically modulated claustrum activity using Designer Receptors Exclusively Activated by Designer Drugs (DREADDs) to explore the impact of altering claustrum activity on global cortical activity. To measure cortical activity and explore resting state and sensory evoked cortical activity in lightly anesthetized mice, we used calcium fluorescence mesoscale imaging in mice that express the fluorescent protein GCaMP6s. Claustrum inhibition via the DREADD HM4Di resulted in increased activity in anterior medial cortical areas, and well as increased areas of local functional connectivity during resting-state activity. An opposite result occurred when the claustrum was excited using the DREADD HM3Dq. Claustrum inhibition also resulted in increased amplitude of cortical response in the visual cortex following visual stimulation, which was the only sensory paradigm to show an effect. These results are consistent with the finding that the claustrum has a large feed forward inhibitory effect on the PFC, and the topographic organization of claustral projections. Manipulating claustrum activity also resulted in changes in functional correlation across cortical regions, resulting in increased decorrelation between anterior medial ROIs and lateral and posterior ROIs following claustrum inhibition, as well as changes in local areas of functional connectivity. These data support the hypothesis that the claustrum has a role in mediating cortical activity during both resting and sensory evoked states.

Disclosures: **R. Zahacy:** None. **Y. Ma:** None. **I.R. Winship:** None. **J. Jackson:** None. **A.W. Chan:** None.

Poster

PSTR126. Cortical Network Dynamics

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR126.27/D15

Topic: B.07. Network Interactions

Support: FRM – ALZ201912009643
ANR Hippocomp: ANR-21-CE37-0011

Title: Rhythmic sensory stimulation to restore brain dynamics and boost memory in a new mouse model of early Alzheimer's disease

Authors: *M. AGUILERA[1,2], S. CASTRO[3,4], D. BATTAGLIA[5,4], R. GOUTAGNY[2,1]; [1]Univ. de Strasbourg, Strasbourg, France; [2]CNRS UMR7364, Strasbourg, France; [3]Aix-Marseille Univ., Marseille, France; [4]USIAS, Strasbourg, France; [5]INS, Aix-Marseille Univ., Marseille, France

Abstract: Alzheimer's disease (AD) is a neurodegenerative pathology characterized by a progressive and irreversible deterioration of cognitive functions, especially memory. A critical challenge in current research on AD is to find a reliable and early marker of the disease. Recently, our team showed that the new humanized *App^{NL-F}xMAPT* double knock-in (dKI) mouse model could generates key information on the initial disease's stage. To better characterize network alteration in the early stage of the disease, we performed long-term high-density EEG recording starting at 2-month of age in dKI and control littermate mice undergoing behavioral testing. Using EEG microstates dynamic analyses, our preliminary results show a decrease in microstates transition together with a reduced microstates sequences complexity starting at 4-months in dKI mice. Strikingly, these microstates dynamic alterations were concomitant with the emergence of the first recognition deficits in an object-place association task. This prompt us to test the effect of a non-invasive visual stimulation on EEG microstates dynamic as recent studies have shown that 40 Hz gamma entrainment via visual stimulation protocol (vGENUS) reduce AD pathology (both at the behavioral and histopathological levels). We showed that vGENUS treatment effectively rescued memory deficits in the object-place association task as well as EEG microstates dynamic and complexity in dKI mice. Our results therefore provide new insight into early network dynamic alterations and beneficial effect of a non-invasive gamma visual stimulation in AD.

Disclosures: **M. Aguilera:** None. **S. Castro:** None. **D. Battaglia:** None. **R. goutagny:** None.

Poster

PSTR126. Cortical Network Dynamics

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR126.28/D16

Topic: B.07. Network Interactions

Support: EU MSCA-ITN "iConn"
EU-MSCA COFUND - AMIDEX Temporal Networks

Title: States of functional connectivity flow and their multiplex dynamics in human epilepsy and postictal aphasia

Authors: *D. BATTAGLIA[1], C. BENAR[1], A. TREBUCHON[2], A. BARRAT[1], N. PEDRESCHI[3];
[1]Aix-Marseille Univ., Marseille, France; [2]Assistance Publique Hôpitaux Marseille (APHM), Marseille, France; [3]Oxford Univ., Oxford, United Kingdom

Abstract: Cognitive function relies on the flexible update of the way in which neuronal populations coordinate their activity, described by dynamic Functional Connectivity (dFC). Therefore, to characterise a brain state, it is important to describe how networks change in time, flowing across different configurations beyond how networks are structured on average. Specifically, the mechanisms that cause aphasia as a transient post-seizure symptom in epileptic patients are yet unknown. We analyse intracranial EEG (sEEG) recordings of patients suffering from pharmaco-resistant epilepsy with postictal aphasia. We study the Functional Connectivity (FC) between different cortical sites in a time- and frequency-resolved manner, representing each recording as a time-varying, multilayer network (dynamic multiplex). We studied in particular: the rate of overall reconfiguration of links from one frame to the next, or dynamic Functional Connectivity (dFC) speed; and the stability of network modules through time, by means of a dynamic modular Allegiance (dA) analysis. The combination of these two approaches allows identifying states of "Functional Connectivity flow" (beyond connectivity states), defined as epochs in which network reconfiguration occurs with comparable speed and degree of spatio-temporal coordination. In other words, we introduce a framework to define different "styles" of network dynamics with faster or slower and more organised or more random flows of reconfiguration.
Our unsupervised analyses reveal then that high-frequency dFC is slowed down in a long postictal phase lasting well beyond the ictal episodes themselves. Furthermore, a pathological state of slow and poorly structured network flow consistently co-occurs with episodes of aphasia symptoms annotated by the clinicians. In conclusion, our multiplex network dynamics description cast light on functional mechanisms of postictal cognitive dysfunction at the level of individual patients.

Disclosures: D. Battaglia: None. **C. Benar:** None. **A. Trebuchon:** None. **A. Barrat:** None. **N. Pedreschi:** None.

Poster

PSTR126. Cortical Network Dynamics

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR126.29/D17

Topic: B.07. Network Interactions

Support: NIH NINDS K23NS112339

Title: Multiple timescale feedback stabilizes recurrent neural networks and promotes multiscale sequence learning

Authors: *B. N. LUNDSTROM[1], T. J. RICHNER[2];
[2]Mayo Clin., [1]Mayo Clin., Rochester, MN

Abstract: Introduction: The brain is a highly connected recurrent neural network that balances excitatory and inhibitory signals. However, positive feedback loops create runaway excitability. Therefore, network connectivity must either be constrained to avoid positive feedback, or feedback mechanisms are required to maintain stability. Spike frequency adaptation (SFA) and short-term synaptic depression (STD) are common features of single neurons and decrease excitability. However, neural networks are often modeled without incorporating SFA and STD. *Methods:* We have developed a framework for understanding multiple timescale feedback. This multiscale adaptation approximates fractional dynamics, which is non-local, history dependent, and implicated in efficient coding and stimulus prediction. Fractional derivative responses contain information about the stimulus history as well as its derivative. Stemming from single neuron and rodent data, we developed filter-based neural mass models and conductance-based models that incorporated this short-term plasticity, including spike frequency adaptation and synaptic depression. *Results:* We find that feedback mechanisms such as SFA and STD reduce runaway excitability, even in randomly connected neural networks. Without any additional tuning of the connectivity, these networks exhibit edge of chaos behavior and therefore encode stimuli for extended periods. Interestingly, SFA can be incorporated directly into the state transition matrix, similar to modifying the connectivity matrix. Multiple timescale SFA and STD have distinct properties that promote network stability but also allow the network to maintain a high degree of flexibility. In addition, due to the mathematical properties of fractional dynamics, multiple timescale adaptation allows for both a slow and fast sequence of inputs to be processed in the same order. In other words, sequence learning can be invariant to presentation frequency. For example, this would allow for the translation of slow learning to be implemented during faster performances. *Conclusion:* Overall, we implement multiple timescale feedback in recurrent neural network models. We show that multiple timescale adaptation improves network stability and can enhance sequence learning.

Disclosures: B.N. Lundstrom: None. **T.J. Richner:** None.

Poster

PSTR127. Astrocyte-Neuron Interactions in Physiology

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR127.01/D18

Topic: B.09. Glial Mechanisms

Support: BBSRC project grants BB/J018422/1 and BB/J017809/1
This project has received funding from the European Union's Horizon 2020 research and innovation programme under the grant agreement NEUROPA No 863214.

Title: Astrocyte Calcium as a homeostatic switch for barrel cortex experience dependent synaptic plasticity.

Authors: N. N. NGUM[1], A. H. BAZZARI[1], F. DELICATA[1], A. LUDLAM[1], E. J. HILL[1,2], S. GLAZEWSKI[3], ***R. PARRI**[1];
[1]Col. of Hlth. and Life Sci., Aston Univ., Birmingham, United Kingdom; [2]Chem., Loughborough Univ., Loghborough, United Kingdom; [3]Univ. of Keele, Univ. of Keele, Keele, United Kingdom

Abstract: In rodents, whisker deprivation induces changes in neuronal responses in the somatosensory barrel cortex. These experience dependent plasticity (EDP) changes are mediated by two forms of cortical plasticity: Hebbian, and Homeostatic synaptic and cellular changes. In response to several days of unilateral all-whisker sensory deprivation of one side of the snout, a PKA-dependent form of long term potentiation (LTP) emerges at L4-L2/3 synapses in addition to the NMDA receptor dependent LTP observed in undeprived animals. We found that a PKA-dependent LTP mechanism was also present in an astrocyte IP_3R2 genetic deletion mouse indicating an astrocytic involvement. To reduce astrocyte cytoplasmic calcium signaling we selectively overexpressed a plasma membrane calcium extrusion pump CalEx by AAV transfection in barrel cortex astrocytes. This again mimicked the effect of whisker deprivation in inducing the emergence of a PKA dependent LTP mechanism but in whisker intact animals. These findings suggest that the emergence of L4 to L2/3 PKA-dependent LTP expression mechanism is a homeostatic response to whisker deprivation, and that a reduction in astrocyte calcium signalling is sufficient to induce this response, so placing astrocytes as gatekeepers of whisker sensory experience- dependent plasticity.

Disclosures: N.N. Ngum: None. **A.H. Bazzari:** None. **F. Delicata:** None. **A. Ludlam:** None. **E.J. Hill:** None. **S. Glazewski:** None. **R. Parri:** None.

Poster

PSTR127. Astrocyte-Neuron Interactions in Physiology

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR127.02/D19

Topic: B.09. Glial Mechanisms

Support: NSF Research Initiation Award (HRD 1401026)
NSF IOS Neural Systems Awards (IOS 1755341 and 1755033)

Title: How astrocytic modulation impacts neuronal network development?

Authors: *V. A. N. TALABATTULA[1], M. MOORE[2], R. DZAKPASU[3], M. K. TEMBURNI[1]; [2]OSCAR Imaging Facility, [1]Delaware State Univ., Dover, DE; [3]Dept. of Physics and Dept. of Pharmacol. and Physiol., Georgetown Univ., Washington, DC

Abstract: How astrocytic modulation impacts neuronal network development?
Synchronous oscillations are necessary for establishing functional neuronal networks in normal vertebrate brain development - however, the mechanisms of neuronal synchronization are not fully understood. Existing models of synchronous activity assume that it is intrinsic to neurons. Astrocytes have been shown to modulate oscillatory activity in networks of neurons possibly by releasing gliotransmitters like glutamate and ATP. We have established pure and mixed (astrocyte and neuronal) cultures from the optic tectum of the developing chicken brain and recorded neuronal network activity using three different the multi-electrode array systems, MED64, Axion and Multichannel Systems. Our preliminary results suggests that astrocytes are necessary for synchronous activity of neurons in culture and the reintroduction of astrocytes retrieved the neuronal spiking within *in vitro* cultures.
To further dissect the molecular pathways involved, we targeted the astrocytic metabotropic glutamate receptor (mGluR) pathway as a mechanism by which astrocytes influence synchronous firing. Astrocytes express mGluRs that consist of the same subunits and stoichiometry as those expressed in neurons. To test this model, we expressed the calcium sensor GCaMP6F to assay Ca^{++}activity and a truncated mGluR subunit (mGluR DN) which acts as a dominant negative by blocking downstream signaling of the mGluR1 pathway using the lentiviral vector pUltrahot in primary chick astrocytes. Preliminary results show that astrocytes expressing the mGluR DN have reduced calcium elevation upon mGluR stimulation. We will co-culture these mGluR DN astrocytes with neurons on MEAs to test for the development of synchrony.Lastly, we will take a pharmacological approach and add the mGluR1 antagonist, A841720, to neuron-astrocyte co-cultures and pure neuronal cultures. Synchronous activity will be recorded using the MCS MEA system and network activity parameters such as synchrony index (SI), spike amplitude and spike rates will be determined. We expect that the mGluR1 antagonist treated cultures will have a reduced Synchrony Index.This study is supported by NSF Research Initiation Award (HRD 1401026) and NSF IOS Neural Systems Awards (IOS 1755341 and 1755033)

Disclosures: V.A.N. Talabattula: None. **M. Moore:** None. **R. Dzakpasu:** None. **M.K. Temburni:** None.

Poster

PSTR127. Astrocyte-Neuron Interactions in Physiology

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR127.03/D20

Topic: B.09. Glial Mechanisms

Support: NSF 1707356
NSF 2014862
NIH award 5R01MH095980-08

Title: Role of perisynaptic astroglia during plasticity in a synaptic cluster

Authors: *A. J. NAM[1], M. KUWAJIMA[2], J. M. MENDENHALL[2], P. H. PARKER[2], D. D. HUBBARD[2], D. C. HANKA[2], W. C. ABRAHAM[3], K. M. HARRIS[2];
[1]Neurosci., Univ. of Texas At Austin Inst. For Neurosci., Austin, TX; [2]Ctr. for Learning & Memory, Univ. of Texas At Austin, Austin, TX; [3]Univ. of Otago, Dunedin, New Zealand

Abstract: Astrocytes are intimately involved in vital processes at a single synapse, from formation to maintenance to removal. Astrocytes occupy largely non-overlapping domains, and perisynaptic astrocytic processes (PAPs) interdigitate the dense neuropil of dendritic spines and axons, where they influence information processing. Synapse clusters comprise neighboring synapses along a single dendritic segment that share resources. Clustered synapses are more effective in initiating an action potential than the same synapses distributed across multiple dendrites. Astrocyte calcium elevations that modulate neuronal activity may influence coordinated activation of synapses within a cluster. However, the role that astrocytes play in modulating synapse clusters during plasticity remains an open question. Here we leveraged 3D reconstruction from serial section electron microscopy (3DEM) and machine learning to investigate whether the presence of PAPs enhances synaptic plasticity in a cluster. An automated pipeline was developed to explore PAP morphological changes 30 minutes and 2 hours following the induction of long-term potentiation (LTP) and concurrent long-term depression (cLTD), widely accepted cellular mechanisms of learning and memory. LTP was induced in the middle molecular layer (MML) of dentate gyrus of adult rats *in vivo* via delta-burst stimulation of the medial perforant pathway. cLTD occurred simultaneously in the outer molecular layer (OML). The contralateral hemispheres received only baseline stimulation to the medial path and served as within-animal controls. Serial section electron microscopy was used to capture high resolution images of the dentate gyrus neuropil. Preliminary analyses revealed that over 70% of synapses in the dentate gyrus had an astrocytic process at the axon-spine interface. In addition, the length of the axon-spine interface with astrocytic coverage scaled linearly with synapse size, thus maintaining proportional availability of astrocytic resources. The relationship between astrocytic contact length and individual synapse size was expanded in parallel with synapse growth during LTP. Moreover, the closer synapses were to one another, the more similar they were in size during both control and LTP conditions. These early results suggest astrocytic processes respond to or coordinate changes in synapse size in synaptic clusters along dendrites.

Disclosures: **A.J. Nam:** None. **M. Kuwajima:** None. **J.M. Mendenhall:** None. **P.H. Parker:** None. **D.D. Hubbard:** None. **D.C. Hanka:** None. **W.C. Abraham:** None. **K.M. Harris:** None.

Poster

PSTR127. Astrocyte-Neuron Interactions in Physiology

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR127.04/D21

Topic: B.09. Glial Mechanisms

Support: R01AG065836
R01NS036692
R01NS123069

Title: Perineuronal Nets restrict pericellular glial coverage and facilitate astrocytic ion and glutamate homeostasis at tripartite synapses

Authors: *B. P. TEWARI[1], A. WOO[1], C. PRIM[1], L. CHAUNSALI[1], I. F. KIMBROUGH[1], K. J. ENGEL[2], J. L. BROWNING[2], S. L. CAMPBELL[3], H. SONTHEIMER[1];
[1]Neurosci., Univ. of Virginia, Charlottesville, VA; [2]Sch. of Neurosci., [3]Animal and Poultry Sci., Virginia Tech., Blacksburg, VA

Abstract: Perineuronal nets (PNNs) are lattice-like coatings of highly organized extracellular matrix consisting of mainly hyaluronic acid, chondroitin sulfate proteoglycans (CSPGs), tenascins, and link proteins. PNN condensation marks the closure of the critical period of neuroplasticity in several brain areas thereby suggesting a role of PNN in stabilizing synaptic contacts to prevent synaptic plasticity. Sulfated proteoglycans equip PNNs with a high density of stationary negative charges owing to which PNNs influence the ionic homeostasis and excitability of their enclosed neurons. Since PNN coats the entire cell surface sparing only the lattice holes, we hypothesized that lattice holes must be preferred sites of synapses. Using high-resolution 3-D confocal imaging we indeed found that essentially all PNN holes contain either excitatory or inhibitory synapses and some contained both. Importantly a vast majority of PNN holes (>75%) also contained astrocytic processes making them tripartite synapses. The astrocytic processes in the PNN holes expressed proteins tasked with astrocytic Glu (EAAT2) and K^+ homeostasis (Kir4.1). During development, PNNs mature concurrently with astrocytes, leading to progressively reducing the astrocytic coverage on PNN-enclosed neurons confined to PNN holes. To ascertain the functional significance of PNN holes harboring tripartite synapses we employed enzymatic or genetic means to disrupt PNNs. Both approaches caused an increase in astrocytic coverage of the neuronal membrane without altering synaptic contacts. Furthermore, whole-cell patch-clamp recordings were used to measure the astrocytic uptake of synaptically released glutamate and K^+. PNN disruption reduced the glutamate and potassium uptake by astrocytes suggesting that holes in the PNNs serve as a container that channels synaptically released glutamate and depolarization-released K+ ions towards astrocytic processes to prevent spillage to extrasynaptic sites. These findings reveal a hitherto unrecognized synergy between astrocytes and PNNs that optimizes astrocytic homeostatic functions on inhibitory PNN-expressing neurons.

Disclosures: B.P. Tewari: None. **A. Woo:** None. **C. Prim:** None. **L. Chaunsali:** None. **I.F. Kimbrough:** None. **K.J. Engel:** None. **J.L. Browning:** None. **S.L. Campbell:** None. **H. Sontheimer:** None.

Poster

PSTR127. Astrocyte-Neuron Interactions in Physiology

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR127.05/D22

Topic: B.09. Glial Mechanisms

Support: Coordenação de Aperfeiçoamento de Pessoal de Nível Superior (CAPES)
Programa de Excelência Acadêmica (PROEX-CAPES)
Fundação de Apoio ao Ensino, Pesquisa e Assistência of the Hospital das Clínicas, Faculty of Ribeirão Preto, University of São Paulo (FAEPA)
Fundação de Amparo à Pesquisa do Estado de São Paulo (FAPESP)

Title: Diversity of responses of astrocytes from the nucleus of the solitary tract to glutamate application

Authors: *M. MALERBA DE SOUZA[1], L. RAKAUSKAS ZACHARIAS[2], R. M. LEAO[3];
[1]Physiol., [2]Physiology/Neural and Behavioral Sci., Univ. of São Paulo, Ribeirão Preto, Brazil;
[3]Physiol., Univ. of Sao Paulo, Ribeirao Preto, Brazil

Abstract: Astrocytes play essential functions modulating several physiological responses. They make close contacts with the pre- and postsynaptic sites and actively modulate synaptic transmission, sensing the released neurotransmitter, and in response release gliotransmitters which modulates neuronal function. The nucleus of the solitary tract (NTS) is a brainstem area that receives information from the periphery mainly via the vagus nerve, forming the solitary tract (ST). The NTS mediates several cardiovascular, respiratory, digestive and metabolic functions and reflexes and NTS astrocytes modulate several physiological reflexes and neurotransmission from the solitary tract. Although the influence of astrocytes on glutamatergic neurotransmission in the NTS has been well reported, how NTS astrocytes respond to glutamate is not well known. Here we analyze the activation profile of NTS astrocytes by glutamate in the subpostremal NTS region, using Ca^{2+} imaging. We loaded subpostremal NTS astrocytes with the calcium sensor Fluo-4-AM and measured their response to glutamate by changes in cytosolic calcium. We show most NTS astrocytes (61%) responded quickly to the perfusion of L-glutamate (500 µM; n = 7 animals; 15 slices; 170 cells) increasing cytosolic calcium, while a minor fraction responded with a decrease in calcium (31%) and the remaining cells were not responsive to glutamate (8%). The response of glutamate is strongly reduced by applying the AMPA/kainate antagonist DNQX (10 µM; n = 4 animals; 8 slices; 95 cells), showing that it is primarily dependent on the activation of these receptors. Here, 46% of astrocytes increased their activity, 46% reduced, and 8% did not change their cytoplasmic calcium. Additionally, it is attenuated in the presence of the action potential blocker tetrodotoxin (TTX - 500 µM; n= 8 animals; 14 slices; 136 cells), showing that neurons amplify the response of the astrocytes, possibly by releasing more glutamate. Here, 42% of astrocytes responded positively to glutamate, 50% negatively, and 8% did not respond. Our results showed that NTS astrocytes express a heterogeneous response to glutamate, with glutamate-activated and inhibited cells, and probably participating in the gliotransmission modulating the physiological responses mediated by the NTS.

Disclosures: **M. Malerba de Souza:** None. **L. Rakauskas Zacharias:** None. **R.M. Leao:** None.

Poster

PSTR127. Astrocyte-Neuron Interactions in Physiology

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR127.06/D23

Topic: B.09. Glial Mechanisms

Support: R01MH 129797
Leon Levy Scholarship in Neuroscience

Title: Astrocytic RNA regulation impacts synaptic function and animal behavior by modulating G-protein signaling and phagocytosis

Authors: ***P. J. LITUMA**[1], E. BARRIO-ALONSO[1], N. WAYLAND[1], K. LYSEK[1], D. COLAK[1], K. TAN[2], M. F. WILKINSON[2], P. E. CASTILLO[3];
[1]Brain and Mind Res. Inst., Weill Cornell Med., New York, NY; [2]Dept. of Obstetrics, Gynecology, and Reproductive Sci., Sch. of Med. Univ. of California San Diego, San Diego, CA; [3]Dominick P. Purpura Dept. of Neurosci., Albert Einstein Col. of Med., Bronx, NY

Abstract: Astrocytes are critical components of neural circuits that are involved in shaping animal behavior and contribute to brain disorders. While astrocytes serve diverse roles, including ion homeostasis, neurotransmitter clearance, synapse remodeling, and synaptic modulation the mechanisms that regulate the diverse biological functions of these complex cells are ill-defined. Despite being the only RNA regulatory pathway that is associated with multiple mental illnesses, the nonsense-mediated mRNA decay (NMD) pathway presents an unexplored regulatory mechanism for astrocyte function in the central nervous system. To address this knowledge gap, we leverage mouse genetic strategies to delete *Upf2*, a specific component of the NMD machinery, in astrocytes of adult (2 months) male and female mice. *Upf2* conditional deletion led to avoidance behavior in mice. The behavioral phenotype was accompanied with aberrant synaptic transmission and long-term synaptic plasticity in hippocampus. Our data indicate astrocytic NMD deficiency alters molecular pathways that influence neuronal function and ultimately, animal behavior. By characterizing the role of NMD in astrocytes we highlight the underappreciated impact of glial RNA biology on brain function.

Disclosures: **P.J. Lituma:** None. **E. Barrio-Alonso:** None. **N. Wayland:** None. **K. Lysek:** None. **D. Colak:** None. **K. Tan:** None. **M.F. Wilkinson:** None. **P.E. Castillo:** None.

Poster

PSTR127. Astrocyte-Neuron Interactions in Physiology

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR127.07/D24

Topic: B.09. Glial Mechanisms

Support: NINDS NS104478
NINDS NS12619

Title: Neuronal activity drives spatially and temporally heterogeneous astrocyte depolarizations.

Authors: *M. ARMBRUSTER[1], C. DULLA[2];
[1]Tufts Univ., Boston, MA; [2]Tufts Univ. Sch. of Med., Boston, MA

Abstract: Astrocytes are glial cells that interact with neuronal synapses via their distal processes, where they remove glutamate and potassium (K+) from the extracellular space following neuronal activity. Using astrocytic genetically encoded fluorescence voltage imaging, we have previously shown that neuronal activity can drive large, rapid, focal and pathway-specific depolarizations in peripheral astrocyte processes which are driven by neuronally released K+ and glutamate transporter currents. We expand upon this work show to that depolarizations are not limited to excitatory synapses. Neuronal activity also induced areas of astrocytic hyperpolarizations. Additionally, we show that astrocytic depolarizations in peripheral astrocyte processes spatially propagate in a Kir4.1 dependent manner. Lastly, using a low Mg2+ in-vitro slice model of seizures, we show disconnects between neuronal activity and astrocytic voltage responses. These show a several classes of heterogeneous astrocytic voltage changes in response to neuronal activity and raises new questions about the molecular mechanisms controlling this and downstream effects of the depolarizations.

Disclosures: M. Armbruster: None. **C. Dulla:** None.

Poster

PSTR127. Astrocyte-Neuron Interactions in Physiology

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR127.08/D25

Topic: B.09. Glial Mechanisms

Support: Chan Zuckerberg Initiative
L.I.F.E. award
NEI F32EY033629
5R01NS105742-05

Title: Astrocyte CCN1 stabilizes visual circuits

Authors: *L. SANCHO FERNANDEZ[1], M. M. BOISVERT[1], T. DAWOODTABAR[2], J. BURGADO[2], N. J. ALLEN[1];
[1]Salk Inst. for Biol. Studies, La Jolla, CA; [2]UCSD, La Jolla, CA

Abstract: Sensory circuits typically have higher plasticity at younger ages, during which circuit refinement and maturation occurs, and plasticity declines with maturation. We are interested in the mechanisms underlying these differing plasticity levels and how they serve to maintain and stabilize the properties of sensory circuits. Astrocytes play a crucial role in regulating the structure, physiology, and plasticity of synapses, and we hypothesize that astrocytes are actively involved in permitting or restricting plasticity throughout different stages of life. We investigate this in the mouse visual cortex (VC), which has stereotyped periods of high plasticity early in development and reduced plasticity in adulthood. We performed transcriptomic analyses of VC astrocytes and identified CCN1 as a potential "anti-plasticity" factor whose expression increases throughout development into adulthood and decreases in paradigms that induce plasticity. CCN1 (cellular communication network factor 1) encodes a 4-domain secreted protein that can bind to heparan sulfate proteoglycans, integrin receptors, connexin 43, as well as the extracellular matrix. We hypothesize that astrocyte-specific CCN1 promotes the stabilization of neuronal synapses and circuits and actively restricts plasticity in adulthood. Overexpressing CCN1 at a time when its expression is low and plasticity is high (the visual critical period) reduces binocular zone remodeling after monocular deprivation (MD). Whole-cell patch-clamp recordings from neurons in layer 2/3 shows overexpressing CCN1 increases excitatory drive onto fast-spiking inhibitory cells, suggesting that CCN1 promotes the maturation of inhibitory circuits in VC. Knocking out CCN1 (cKO) from adult astrocytes enables remodeling after MD in adult mice. Perineuronal net (PNN) density around inhibitory interneurons is decreased in adult CCN1 cKO mice at baseline and further decreased after MD, indicating that CCN1 acts to stabilize inhibitory circuits. *In vivo* two-photon imaging in adult VC shows that CCN1 cKO results in aberrant binocular circuits, reflecting altered ocular dominance and neuronal connectivity. We have identified CCN1 as a novel astrocyte factor that stabilizes neural circuits.

Disclosures: L. Sancho Fernandez: None. **M.M. Boisvert:** None. **T. Dawoodtabar:** None. **J. Burgado:** None. **N.J. Allen:** None.

Poster

PSTR127. Astrocyte-Neuron Interactions in Physiology

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR127.09/D26

Topic: B.09. Glial Mechanisms

Support: JSPS Grant-in-Aid JP21H00220
JSPS Grant-in-Aid JP21H02588
JSPS Grant-in-Aid JP21H05640
JSPS Grant-in-Aid JP23H04179
AMED Grant JP22dk0207063

Title: Prefrontal Astrocyte Regulation of Behavioral Flexibility

Authors: *B. WULAER[1], T. ENDO[2], E. SHIGETOMI[3,4], S. KOIZUMI[3,4], J. NAGAI[1];
[1]RIKEN Ctr. for Brain Sci., Wako/Saitama, Japan; [2]Phenovance LLC, Kashiwa/Chiba, Japan; [3]Univ. of Yamanashi, Chuo/Yamanashi, Japan; [4]GLIA Center, Univ. of Yamanashi, Chuo/Yamanashi, Japan

Abstract: Behavioral flexibility, the ability to adapt and adjust actions in response to changing circumstances, is essential for survival and goal achievement in a dynamic environment. While the prefrontal cortex plays a central role in regulating behavioral flexibility, the contribution of astrocytes in this process remains unexplored. In this study, we aimed to characterize how prefrontal astrocytes contribute to behavioral flexibility using a combination of experimental techniques including Ca^{2+} imaging, pharmacology, electrophysiology, chemogenetics, behavioral tests, and RNA-sequencing. Initially, we examined if and how prefrontal astrocytes respond to various neurotransmitters. We found that the Ca^{2+} responses of prefrontal astrocytes were distinct between neurotransmitters, and that most profound signaling occurred through the activation of Gq-coupled alpha1-adrenoceptors. To investigate the effects of chemogenetic stimulation of astrocyte-specific Gq/Ca^{2+} signaling on mouse behavior, we conducted an extensive battery of 17 behavioral tests. Our results demonstrated that such stimulation induced behavioral inflexibility in mice, while it spared locomotion, motor coordination, working memory, and novelty recognition. To reveal underlying mechanisms of how chemogenetic astrocyte Gq stimulation induces behavioral inflexibility, we assessed astrocyte responses to neurotransmitters, neural activity, and transcriptomic changes with astrocyte Gq signaling activation. Our data revealed that the chemogenetic astrocyte stimulation blunted their responses to neurotransmitters. We also observed reduced activity of local CaMKII-expressing excitatory neurons. We will report RNA-sequencing analyses in which we aim to unravel the astrocyte-mediated molecular mechanisms underlying the contribution of astrocytes to behavioral flexibility. Overall, our findings so far uncovered a critical new role for prefrontal astrocytes in regulating behavioral flexibility in a dynamic environment, which may highlight the potential of developing novel treatment strategies focused on astrocytes for disorders related to behavioral flexibility.

Disclosures: **B. Wulaer:** None. **T. Endo:** None. **E. Shigetomi:** None. **S. Koizumi:** None. **J. Nagai:** None.

Poster

PSTR127. Astrocyte-Neuron Interactions in Physiology

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR127.10/D27

Topic: B.09. Glial Mechanisms

Support: NIH Grant F32 NS117776
CZI

Title: Dysregulation of astrocyte-secreted pleiotrophin contributes to neuronal dendrite and synaptic phenotypes in Down Syndrome

Authors: *A. BRANDEBURA[1], M. MICAEL[2], Q. ASBELL[1], N. ALLEN[1];
[1]Salk Inst., San Diego, CA; [2]Univ. of California San Diego, San Diego, CA

Abstract: Astrocytes have come to the forefront as vital players in the regulation of neuronal morphology and synaptic connectivity. A variety of astrocyte-secreted proteins have been shown to act as neurite outgrowth factors, as well as to promote and maintain synaptic homeostasis. During brain development, imbalances in the outgrowth and branching of neuronal processes and alterations to synaptic density can manifest as a variety of neurodevelopmental disorders. Down Syndrome (DS) is one such neurodevelopmental disorder accompanied with intellectual disability. A recent study demonstrated that astrocyte protein secretion is highly dysregulated in cultured cortical astrocytes isolated from the Ts65Dn Mutant (Mut) mouse model of DS, which potentially implicates astrocytes in the etiology of DS. This dataset revealed that secretion of the astrocyte-enriched protein, pleiotrophin (Ptn), is >4-fold reduced from DS astrocytes, leading to the hypothesis that reduced Ptn secretion may contribute to neuronal dendrite and synaptic phenotypes in DS. We characterized Ptn knockout (KO) mice to identify phenotypes that may be in common with Ts65Dn mice. We used the visual cortex as a model system due to its well-characterized circuitry, and Layer V pyramidal neurons from visual cortex have known dendrite outgrowth and spine density deficits in humans with DS. Postnatal day (P)30 and P120 were chosen for analysis timepoints to investigate circuit development during the critical period and circuit stabilization in adulthood, respectively. Both Ptn KO and Ts65Dn Mut mice had shorter and less complex dendrites at P30. Furthermore, a reduced density of dendritic spines and decreased number of excitatory intracortical synapses was observed in both Ptn KO and Ts65Dn Mut mice at P120. Overall, the Ptn KO mice displayed a high degree of similarity to Ts65Dn Mut mice. Therefore, a viral-mediated Ptn overexpression strategy was used to determine if restoration of Ptn levels can rescue neuronal phenotypes in DS. Ptn overexpression was able to fully rescue dendrite length and complexity in Ts65Dn Mut mice compared to Mut mice treated with control virus. Analysis of spine density and colocalized synapses is ongoing. These findings demonstrate that astrocyte-secreted Ptn has important roles in cortical development, and that decreased astrocyte-secreted Ptn levels in DS at least partially contribute to neuronal phenotypes, highlighting the modulatory role of this glial subtype in the manifestation of neurodevelopmental disorders.

Disclosures: **A. Brandebura:** None. **M. Micael:** None. **Q. Asbell:** None. **N. Allen:** None.

Poster

PSTR127. Astrocyte-Neuron Interactions in Physiology

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR127.11/D28

Topic: B.09. Glial Mechanisms

Title: Dissecting neuron-astrocyte crosstalk via an inducible neuronal silencing tool

Authors: *M. FERNÁNDEZ DE LA PUEBLA[1,2], X. ZHANG[3], K. SINDRE ÅBJØRSBRÅTEN[3], W. TANG[3,2];
[1]UiO, Oslo, Norway; [2]Dept. of Clin. and Mol. Med., Norwegian Univ. of Sci. and Technol. (NTNU), Trondheim, Norway; [3]Inst. of Basic Med. Sciences,University of Oslo, Oslo, Norway

Abstract: Astrocytes are an abundant glial cell type in the central nervous system (CNS). Besides being the primary homeostatic and supportive cell type in CNS, a growing body of evidence positions astrocytes as essential players in synaptic plasticity and shaping animal behaviour together with neurons. Astrocytic Ca^{2+} signalling plays a crucial role in the communication between neurons and astrocytes. They are influenced by neurotransmitters released during neuronal stimulation, as well as various neuromodulators released from brain modulatory system, such as noradrenaline and acetylcholine. However, one of the inherent challenges in studying astrocytic Ca^{2+} responses related to neuronal input in awake-behaving animals is the potential influence of simultaneous brain arousal states. Currently, many *in vivo* studies still use harsh stimulation protocols (forced locomotion, air-puff whisker stimulation, electric shock etc.) that innately increase the brain arousal state of the animal, thus masking the possible astrocyte response to neuronal stimulation. Here, we utilized the recombinant adeno-associated virus (rAAV) gene delivery approach to introduce genetically encoded calcium indicators (GECIs) in both neurons and astrocytes, as well as a chemogenetic neuronal silencing tool (a mutant glycine receptor $GlyR\alpha1^{AG}$) in somatosensory cortical layer 2 neurons. Using two-photon Ca^{2+} imaging in head-fixed awake behaving mice, we simultaneously observed activities from somatosensory cortical layer 2 neurons and astrocytes during animal whisker behaviour. With and without activation of chemogenetic silencing of neurons, activities of both neurons and astrocytes are monitored in various behavioural statuses. With this experimental setup, we observed Ca^{2+} elevation in both neurons and astrocytes upon voluntary running behaviour, heightened during the whisker stimulation period. Following the induction of somatosensory neuronal silencing, neuronal Ca^{2+} activity was greatly reduced while astrocytic Ca^{2+} activity was less affected, indicating other significant mechanisms, such as neuromodulatory systems, driving astrocytic Ca^{2+} elevation besides local neuronal input.
Keywords: neuron-astrocyte crosstalk, calcium imaging, somatosensory cortex, neuronal silencing

Disclosures: M. Fernández De La Puebla: None. **X. Zhang:** None. **K. Sindre Åbjørsbråten:** None. **W. Tang:** None.

Poster

PSTR127. Astrocyte-Neuron Interactions in Physiology

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR127.12/D29

Topic: B.09. Glial Mechanisms

Support: Sree Ramakrishna Paramhamsa Research Grant (2020) from Sree Padmavathi Venkateswara Foundation (SreePVF)
Department of Atomic Energy, Government of India, Grant RTI4003

Title: Chronic postnatal chemogenetic activation of CaMKIIα-positive forebrain excitatory neurons modulates adult glial function and metabolism

Authors: *A. PRADHAN[1], S. PATI[1], K. SABA[2,3], P. TIWARI[1], D. KAPRI[1], A. BALAKRISHNAN[1], P. R. CHAUDHARI[1], A. B. PATEL[2,3], V. A. VAIDYA[1];
[1]Dept. of Biol. Sci., Tata Inst. of Fundamental Res., Mumbai, India; [2]Council of Scientific and Industrial Research-Centre for Cell. and Mol. Biol., Hyderabad, India; [3]Acad. of Scientific and Innovative Res. (AcSIR), Ghaziabad, India

Abstract: Early adversity increases vulnerability to developing anxiodepressive behaviors in adulthood. Across multiple pre-clinical models of early adversity, there are reports of glial dysfunction and disrupted amino acid neurotransmission, phenocopying some of the changes noted in patients suffering from major depression. We recently showed that postnatally enhancing Gq signaling in the forebrain excitatory neurons can profoundly impact the emergence of anxiodepressive behaviors and perturb sensorimotor gating, accompanied by altered neuronal glutamate and GABA metabolism in mouse models. Given prior evidence that glial dysfunction is associated with mood disorders, we hypothesized that postnatally enhancing Gq signaling in the forebrain excitatory neurons may also impact glial function in adulthood. To address this, we used a CaMKIIα-tTA::TetO-hM3Dq bigenic mouse line, wherein hM3Dq-DREADD is expressed in CaMKIIα-positive forebrain excitatory neurons, and administered the DREADD agonist, clozapine-N-oxide, or vehicle from postnatal day 2 to 14 to investigate its impact on glial function and metabolism in adulthood. We show that postnatal hM3Dq-mediated activation of forebrain excitatory neurons increases anxiety-like behavior in a task-dependent manner. We also noted an overall bidirectional transcriptional regulation of multiple glia-associated markers, *viz.* GFAP, ALDH1L1, S100β, EAATs, and GATs, as evidenced by a significant decrease and increase in the relative gene expression of these markers within the neocortex and the hippocampus, respectively. We noted a similar bidirectional trend in GFAP and S100β protein levels in these regions. However, postnatally enhancing hM3Dq-mediated activation of CaMKIIα-positive neurons did not alter astrocyte cell density in both the neocortex and the hippocampus. We further investigated the effects of chronic chemogenetic activation of forebrain excitatory neurons on amino acid neurotransmission using ^{1}H-[^{13}C]-NMR spectroscopy and observed a significant decline in glutamate and GABA neurotransmitter turnover and overall astroglial metabolic flux within the neocortex and the hippocampus in adulthood. We are currently addressing the impact of enhanced postnatal Gq signaling in CaMKIIα-positive neurons on astroglial mitochondrial respiration in the forebrain. Our findings indicate that chemogenetically driving Gq signaling transiently during the postnatal window in forebrain excitatory neurons results in enhanced anxiety-like behaviors in adulthood, associated with disrupted glial function and metabolism, a feature that is implicated in the pathogenesis of mood disorders.

Disclosures: **A. Pradhan:** A. Employment/Salary (full or part-time); Tata Institute of Fundamental Research, Mumbai, India. **S. Pati:** None. **K. Saba:** None. **P. Tiwari:** None. **D. Kapri:** None. **A. Balakrishnan:** None. **P.R. Chaudhari:** None. **A.B. Patel:** None. **V.A. Vaidya:** None.

Poster

PSTR127. Astrocyte-Neuron Interactions in Physiology

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR127.13/D30

Topic: B.09. Glial Mechanisms

Support: NIH NINDS R21 NS116664
NIH R01 NS096100

Title: Sonic hedgehog signaling is stimulated in an activity-dependent manner

Authors: *A. LE[1], S. HILL[1], M. FU[1], J. SAUNDERS[1], J. MELL[2], A. R. GARCIA[1];
[1]Biol., [2]Microbiology & Immunol., Drexel Univ., Philadelphia, PA

Abstract: In this study, we investigated whether neural activity modulates Sonic hedgehog (Shh) signaling between neurons and astrocytes. The Shh signaling pathway is a defining molecular signaling pathway in neural development where it exerts regulatory control over patterning, cell fate specification, axon pathfinding, and acts as a powerful mitogen regulating proliferation of neural progenitor cells. In contrast, Shh signaling in the postnatal and adult CNS mediates neuron-astrocyte communication and a growing body of evidence points to its role in regulating astrocyte modulation of synapses. Selective perturbation of Shh signaling in astrocytes leads to elevated synapse number in cortical neurons and an increase in neuronal excitability. Shh is found in axons and studies in slice culture demonstrate that high frequency stimulation induces its release. In this study, we examined whether activity-dependent Shh signaling occurs *in vivo* and further identified Shh-dependent gene expression programs in astrocytes. To explore whether neuronal activity can modulate Shh signaling *in vivo*, we delivered virus carrying DREADD receptors into the cortex of adult mice and monitored Shh activity by expression of *Gli1*, a transcriptional readout of Shh signaling, in $Gli1^{nlacZ/+}$ mice. Chemogenetic activation of cortical neurons increases the number of *Gli1*-expressing astrocytes, demonstrating that neuronal activity stimulates Shh signaling *in vivo*. To investigate the behavioral relevance of activity-dependent Shh signaling, we housed mice in an enriched environment to promote somatosensory activity. We observed a selective increase in *Gli1* expression in the somatosensory, but not the visual cortex, suggesting that experience stimulates Shh signaling in a circuit-specific manner. To identify Shh-dependent genes modulated by experience, we performed RNASeq of cortical astrocytes expressing *Gli1*. We identified over 500 genes that are selectively enriched in these cells compared to all cortical astrocytes. Gene ontology analysis identified 32 synapse-associated genes, including *Hevin* and *Sparc*. Sensory experience modulates *Hevin* and *Sparc* and experience-dependent *Sparc* expression is Shh-dependent. Taken together, our current work revealed a novel activity-dependent role for Shh signaling between neurons and astrocytes and suggests that Shh regulates gene expression programs underlying astrocyte modulation of synapses. Ongoing work aims to determine whether Shh signaling regulates activity-dependent neuronal plasticity.

Disclosures: **A. Le:** None. **S. Hill:** None. **M. Fu:** None. **J. Saunders:** None. **J. Mell:** None. **A. R. Garcia:** None.

Poster

PSTR127. Astrocyte-Neuron Interactions in Physiology

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR127.14/D31

Topic: B.09. Glial Mechanisms

Support: German Research Foundation SPP1757 SA2114/2
Howard Hughes Medical Institute
NIH Grant 1R01NS126043
NIH Grant P30 NS050274
NIH Grant P50 MH084020
NIH Grant 1R01GM128997
NIH Grant U19NS104653
NIH Grant 1R01NS124017
NSF Grant IIS-1912293
NSF GRFP DGE1745303
NSF GRFP DGE2139757
Simons Foundation SCGB 542943SPI

Title: Fast-acting antidepressants modulate glial integration of futility signals in larval zebrafish

Authors: *M. DUQUE RAMIREZ[1], A. CHEN[2], E. HSU[3], S. NARAYAN[4], S. BEGUM[1], G. SAHER[5], A. COHEN[1], D. E. OLSON[6], D. E. BERGLES[7], M. C. FISHMAN[1], F. ENGERT[1], M. B. AHRENS[8];
[1]Harvard Univ., Cambridge, MA; [2]Janelia Res. Campus, Ashburn, VA; [3]Neurobio., Johns Hopkins Univ., Baltimore, MD; [4]Allen Inst., Seattle, WA; [5]Max-Planck Inst. Exptl. Med., Gottingen, Germany; [6]UC Davis, Davis, CA; [7]Johns Hopkins Univ. Sch. Med., BALTIMORE, MD; [8]Janelia Res. Campus / HHMI, Ashburn, VA

Abstract: Rapidly-acting antidepressants like ketamine hold promise for the treatment of major depressive disorder (MDD), despite a limited understanding of their mechanism of action. Fast-acting antidepressants bind various molecular targets and modulate neuronal activity in many brain regions to cause long-lasting behavioral changes. However, connecting the molecular, circuit, and behavioral effects of fast-acting antidepressants has been challenging due to the difficulty of interrogating their effects on brain dynamics during behavior in rodent models. We therefore leveraged the genetic and optical accessibility of larval zebrafish to assess the effects of fast-acting antidepressants on a recently discovered futility-induced passivity behavior as well as their effects on whole-brain neural and astroglial calcium dynamics. Rapid-acting antidepressants with diverse pharmacological targets suppress futility-induced passivity in fish, as they do in rodents. While antidepressants are thought to primarily act on neurons, using brain-

wide imaging in vivo we found that ketamine, but not psychedelics or typical antidepressants, elevates cytosolic calcium in astroglia for many minutes. Chemogenetic and optogenetic perturbations of astroglia revealed that the aftereffects of calcium elevation are sufficient to suppress futility-induced passivity by inhibiting astroglial integration of futile swimming. In vivo two photon imaging in mice revealed that ketamine also profoundly increased cytosolic calcium levels in rodent cortical astrocytes, indicating broad conservation of this phenomenon across species.

Disclosures: **M. Duque Ramirez:** None. **A. Chen:** None. **E. Hsu:** None. **S. Narayan:** None. **S. Begum:** None. **G. Saher:** None. **A. Cohen:** A. Employment/Salary (full or part-time); Q-State Biosciences. E. Ownership Interest (stock, stock options, royalty, receipt of intellectual property rights/patent holder, excluding diversified mutual funds); Q-State Biosciences. **D.E. Olson:** A. Employment/Salary (full or part-time); Delix Therapeutics Inc.. B. Contracted Research/Research Grant (principal investigator for a drug study, collaborator or consultant and pending and current grants). If you are a PI for a drug study, report that research relationship even if those funds come to an institution.; Delix Therapeutics Inc.. C. Other Research Support (receipt of drugs, supplies, equipment or other in-kind support); Delix Therapeutics Inc.. E. Ownership Interest (stock, stock options, royalty, receipt of intellectual property rights/patent holder, excluding diversified mutual funds); Delix Therapeutics Inc.. **D.E. Bergles:** None. **M.C. Fishman:** None. **F. Engert:** None. **M.B. Ahrens:** None.

Poster

PSTR127. Astrocyte-Neuron Interactions in Physiology

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR127.15/D32

Topic: B.09. Glial Mechanisms

Support: NIH Intramural grant 1ZIADK075172-01

Title: Whole animal single cell RNA-sequencing reveals cell non-autonomous tissue-specific changes induced by ER UPR activation in glia.

Authors: *C. A. SHEELER, A. E. FRAKES;
GBB, NIDDK, Rockville, MD

Abstract: The ability to maintain protein homeostasis decreases with age. Previously, we found that a subset of astrocyte-like glial cells regulates organismal protein homeostasis and longevity in C. elegans via the unfolded protein response of the ER (ER UPR). Constitutive activation of the UPRER transcription factor, xbp-1s, in only four astrocyte-like glial cells initiates a robust cell non-autonomous activation of the UPRER in distal cells. This prevents age-onset loss of UPRER, reduces protein aggregation, and extends lifespan. Mutants deficient in neuropeptide processing and secretion suppress glial cell non-autonomous activation of these phenotypes, suggesting that the cell non-autonomous signal is a neuropeptide. (Frakes et al 2020). However,

the downstream effectors mediating ER stress resistance and longevity are unknown. To determine the organism-wide transcriptional changes induced by glial cell non-autonomous signaling, we performed single cell RNAseq on adult glial xbp-1s and control animals (2 samples; ~30k worms/sample/genotype). Cells were barcoded and sequenced on a Chromium 10X and reads aligned with CellRanger 7.1.0. Low-dimensional leiden clustering revealed 22 unique clusters, mapped to 12 distinct tissue types including neurons, glia, and intestine. Pseudobulk analysis of the combined data (DESeq2) identified differentially expressed genes (DEGs) in nearly all tissue populations. Gene ontology enrichment (gProfiler) identified both expected and novel tissue-specific pathway changes in this model. Upregulated DEGs in the intestine drive terms such as proteolysis and lysozyme activity. This recapitulates previous work showing increased macroautophagy and lysosome activation in glial xbp1s animals (Metcalf et al 2022). Interestingly, we find different signatures in other tissues, including carbohydrate biosynthesis and cell-cell signaling which are terms upregulated in muscle of glial xbp1s animals. In conclusion, whole-organism single cell RNAseq shows that xbp-1s driven ER UPR activity in 4 astrocyte-like glia drives extensive cell non-autonomous, tissue-specific changes. Though our previous work focused on ER UPR activation in the intestine, our single cell RNAseq data reveals robust transcriptional changes in distal tissues previously not considered, such as muscle. Future work will elucidate the role of neuropeptide signaling in this transcriptomic effect through assessment of altered receptor-ligand interactions. Lastly, we will confirm the biological impact of these tissue-specific pathways in the glial xbp-1s animals which could identify novel regulators of ER stress resistance and longevity.

Disclosures: C.A. Sheeler: None. **A.E. Frakes:** None.

Poster

PSTR127. Astrocyte-Neuron Interactions in Physiology

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR127.16/D33

Topic: B.09. Glial Mechanisms

Title: Mechanism of NMDA receptor potentiation by lactate

Authors: *H. FIUMELLI[1], G. HERRERA LOPEZ[1], J. GIRGIS[1], F. LEMTIRI-CHLIEH[2], N. CARRANO[3], M. DI LUCA[3], F. GARDONI[3], P. J. MAGISTRETTI[1];
[1]King Abdullah Univ. of Sci. and Technol., Thuwal, Saudi Arabia; [2]Neurosci., Uconn Health, Sch. of Med., Farmington, CT; [3]Univ. of Milano, Milano, Italy

Abstract: Astrocyte-derived lactate fuels high-energy demands of neurons and acts as a signal promoting synaptic plasticity and memory consolidation. Lactate regulates neuronal excitability and modulates gene expression related to synaptic plasticity and neuroprotection but its mode of action is uncertain. Using patch-clamp recordings in cultured cortical neurons, we found that lactate enhances, in a calcium-dependent manner, the amplitude and inactivation time constant of NMDA receptor currents (I_{NMDAR}) evoked by brief applications of glutamate and glycine. These

effects were not observed by agonists of the HCAR1/gpr81 receptor, while monocarboxylate transporters and lactate dehydrogenase inhibitors prevented the lactate-mediated increases in I_{NMDAR} amplitude. Intracellular infusion of specific CaMKII peptide inhibitors also abolished the potentiation of peak I_{NMDAR} responses by lactate, and manipulating the intracellular and extracellular redox balance additively influenced the I_{NMDAR}, indicating the existence of a mechanism that requires the entry of lactate into neurons, redox changes via lactate oxidation to pyruvate, and involvement of CaMKII. Quantitative immunoprecipitation showed that lactate increased the binding of CaMKII to GluN2B, whereas interfering with the binding between CaMKII and GluN2B prevented the potentiation of I_{NMDAR} responses by lactate. Proximity ligation assays between GluN2B and the postsynaptic density marker PSD-95 revealed that lactate induced an accumulation of GluN2B in dendritic spines, an effect that was prevented by a CaMKII peptide inhibitor. Together, these findings establish a mechanistic link between astrocyte-derived lactate and the CaMKII- and NMDAR-dependent synaptic plasticity.

Disclosures: H. Fiumelli: None. **G. Herrera Lopez:** None. **J. Girgis:** None. **F. Lemtiri-Chlieh:** None. **N. Carrano:** None. **M. Di Luca:** None. **F. Gardoni:** None. **P.J. Magistretti:** None.

Poster

PSTR127. Astrocyte-Neuron Interactions in Physiology

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR127.17/D34

Topic: B.09. Glial Mechanisms

Support: NRF Grant

Title: Investigating the mechanisms of glial phagocytosis during motor learning

Authors: *Y.-J. CHOI, W.-S. CHUNG;
Korea Advanced Inst. in Sci. and Technol., Daejeon, Korea, Republic of

Abstract: The formation and elimination of synaptic connections in the brain are dynamically controlled processes that are crucial for normal brain function. While the molecular mechanisms underlying synapse formation have been well-studied, the mechanisms involved in synapse elimination are not yet fully understood. Recent studies have revealed that glia play a key role in mediating synapse elimination by phagocytosing unnecessary synapses during development and in adulthood, thereby contributing to neural circuit maturation and brain homeostasis. However, the upstream regulators that control glia-mediated synapse elimination remain unknown In our lab, we performed chemical screening and discovered that dopamine receptor agonists strongly enhance astrocyte-mediated phagocytosis in vitro, suggesting that neuromodulators such as dopamine may act as upstream regulators of synapse elimination by glial cells. Dopamine is a well-known neuromodulator that is involved in various cognitive functions, including motor control, reinforcement learning, and executive function. It has been shown that dopamine can

modulate synaptic potentiation and remodeling, which are essential for proper learning. Based on this, we hypothesized that glial phagocytosis may be regulated by synaptic potentiation and depotentiation, and that glial function is critical for normal motor learning. Our results show that astrocytic phagocytosis of striatal excitatory synapses is increased during motor learning tasks and that this process is mediated by the astrocytic phagocytosis receptor Megf10. Furthermore, we found that chemogenetic modulation of dopamine secretion enhances astrocytic phagocytosis of corticostriatal presynapses. Interestingly, we observed that astrocytic phagocytosis of striatal excitatory postsynapses is reversed in D1-MSNs and D2-MSNs, suggesting that dopamine-mediated modulation of neural activity in the striatum regulates astrocytic phagocytosis activity of synapses. These findings provide insight into the molecular mechanisms underlying synapse elimination by glia and suggest that modulation of dopamine signaling may be a potential therapeutic target for neurological disorders characterized by disrupted synapse elimination.

Disclosures: Y. Choi: None. **W. Chung:** None.

Poster

PSTR128. Aging-Related Molecular and Cellular Changes

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR128.01/D35

Topic: C.01. Brain Wellness and Aging

Support: T32 GM008216

Title: Histone variant H2BE promotes astrocyte senescence and age-related memory deficits in mice

Authors: *S. LOUZON[1], E. HYATT[1], Q. QIU[1], N. PRESCOTT[2], Y. DAVID[2], H. WU[1], E. KORB[1];
[1]Univ. of Pennsylvania, Philadelphia, PA; [2]Mem. Sloan Kettering, New York, NY

Abstract: A critical mechanism of chromatin regulation is the exchange of canonical histone proteins in nucleosomes with histone variant proteins. A novel histone variant, H2BE, was initially described as being uniquely expressed in the mouse olfactory system. However, we found that H2BE is robustly expressed throughout the mouse brain utilizing an H2BE-specific antibody. We additionally found that H2BE accumulates in astrocytes with age in the brain. Similarly, we discovered that H2BE accumulates in cultured astrocytes as they undergo senescence, a process of irreversible cell cycle arrest and inflammatory signaling that occurs in advanced age and promotes neurodegenerative phenotypes. By comparing astrocytes from WT and H2BE KO mice, we then established that H2BE promotes expression of senescence genes in astrocytes. To interrogate the mechanisms of how H2BE might affect gene expression, we then assessed the impact of H2BE on chromatin. Through both Atomic Force Microscopy and ATAC-Sequencing, we discovered that H2BE promotes increased chromatin accessibility. More specifically, we found that loss of H2BE prevents the senescence-induced increase in

accessibility at the transcription start sites of senescence genes, highlighting a possible mechanism through which H2BE can influence gene expression and the progression of cellular senescence. To then determine if H2BE has a role in governing mouse behavior, we performed a T-maze test of working memory. From this paradigm, we found that loss of H2BE, specifically in aged mice, improves their performance on a T-maze test, indicating that H2BE promotes age-related cognitive decline in mice. Taken together, we have identified a novel role for the histone variant H2BE in regulating astrocyte senescence and age-related memory deficits, which ultimately could lead to new chromatin-based approaches to alleviate neurodegenerative phenotypes.

Disclosures: S. Louzon: None. **E. Hyatt:** None. **Q. Qiu:** None. **N. Prescott:** None. **Y. David:** None. **H. Wu:** None. **E. Korb:** None.

Poster

PSTR128. Aging-Related Molecular and Cellular Changes

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR128.02/D37

Topic: C.01. Brain Wellness and Aging

Support: NIH 1RF1 AG072602-01

Title: Cell-type specific alterations of DNA polymerase kappa in the aging brain

Authors: *M. ABDELMAGEED;
Penn State Col. of Med., Hershey, PA

Abstract: Genomic instability increases with age due to an accumulation of DNA damage and distinct DNA damage response pathways have evolved with a host of distinct polymerases that act via different pathways such as nucleotide excision repair, base excision repair to mend DNA lesions. In contrast, the relatively newly discovered Y-family DNA polymerases can bypass damaged DNA in dividing cells, however, their functions in enduring post-mitotic cell types like neurons are largely unknown, where the non-homologous end joining (NHEJ) pathway has been shown to repair double-stranded DNA breaks. To understand the biological role of Y-family polymerases in maintaining the central nervous system (CNS) genome we studied its expression as function of aging and neurodegeneration. We observed that most Y-family polymerases are expressed extensively in the brain and its expression in sub-cellular compartments are significantly altered as a function of chronological age. We identify that such alterations in neurons are non-uniform across different brain areas perhaps reflecting metabolic demand and demonstrate interacting partners of one member of Y-family polymerase, Polymerase kappa (PolK) to be associated with NHEJ pathway proteins. We further identify the subcellular compartments of PolK sites within the neurons and explore the potential relationship of PolK with cytoplasmic DNA and immune activation.

Disclosures: M. Abdelmageed: None.

Poster

PSTR128. Aging-Related Molecular and Cellular Changes

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR128.03/D38

Topic: C.01. Brain Wellness and Aging

Support: RO1AG037868
P30CA177558

Title: Dorsal vs ventral ex vivo hippocampal transcriptome response to corticosterone in young and aged F344 rats

Authors: *D. R. CRAIG, E. M. BLALOCK;
Pharmacol. & Nutritional Sci., Univ. of Kentucky, Lexington, KY

Abstract: Rationale & Objective: Glucocorticoid (GC) signaling is thought to play a key role in stress' negative impact on brain aging (BA). Several lines of evidence indicate that psychosocial stress and stress hormone exposure accelerates BA. In prior work, using *ex vivo* slice preparations of dorsal (DHIP) and ventral hippocampus (VHIP), we examined the GC-dependent, downstream effector molecule, Sgk1; in this preparation Sgk1 mRNA is increased by both stress and aging. DHIP is thought to play a more prominent role in spatial navigation and short-term memory, while VHIP provides feedback inhibition of the stress response. However, little work has examined transcriptional profiles across the D-V axis in response to GCs with age. Here, an *ex vivo* hippocampal slice preparation was used to examine differential GC-driven transcriptional signaling properties in DHIP and VHIP from young and aged (n = 3/ age) male Fisher 344 rats to test the hypotheses that: *Ex vivo* GC exposure changes mRNA levels in expected directions (e.g., elevated Sgk1, suppressed Tnf), that this effect differs along the D-V axis, and that young tissue will show a greater dynamic response than aged. Methods: HIP slices (350 µm thick) were mapped according to their position on the D-V axis and maintained in oxygenated aCSF at 32°C in four-well interface chambers, and incubated in either 0.1% DMSO (Vehicle control) or 3.5 µM corticosterone (Cort) for 2-hrs. Slices were then removed and RNA was isolated. After QC, whole transcriptome RNA sequencing was performed (Illumina HiSeq) using a dual indexed, paired end, strand-specific read-strategy with 25 million reads per sample. Resulting count data were analyzed for Left vs Right, Dorsal vs Ventral, Young vs Aged, and Control vs Cort effects using DeSeq2 (Left vs. Right showed no significant effects, so were pooled for subsequent analyses). Results & Conclusions: The well-established aging transcriptional profile observed in reproductively intact animals was recapitulated in this preparation. Further, GC exposure increased Sgk1, Depp1, and Ptpdc1 mRNA expression (among others) in all regions of young and aged HIP. The effect was greater in young than aged animals, and, within young animals, was greater in ventral than dorsal HIP. Further, GCs mediated a reduction of Il1a, Il1b and Tnf (among others) in a similar pattern. It is interesting to note that some upregulated aging mRNAs such as Cd74 were unaffected by GCs in young animals, but were substantially increased by GC exposure in aged. Finally, the slice preparation

typically used for electrophysiology, is appropriate for investigating age-related, GC-driven transcriptional profiling.

Disclosures: D.R. Craig: None. **E.M. Blalock:** None.

Poster

PSTR128. Aging-Related Molecular and Cellular Changes

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR128.04/D39

Topic: C.01. Brain Wellness and Aging

Support: F31AG079653

Title: Derailed protein turnover in the aging mammalian brain.

Authors: *N. R. RAO[1], A. UPADHYAY[2], J. N. SAVAS[2];
[1]Neurol., [2]Northwestern Univ., Chicago, IL

Abstract: Efficient protein turnover is essential for cellular homeostasis and organ function. Loss of proteostasis is a hallmark of aging, which culminates as a severe reduction in protein turnover rates. To investigate changes in protein turnover dynamics as a function of age, we performed continuous *in vivo* metabolic stable isotope labeling in mice along the aging continuum. First, we discovered that the brain proteome uniquely experiences dynamic global turnover fluctuations during aging compared to heart and liver tissue. Second, in the brain proteome, global protein turnover trends across aging displayed sex-specific differences that were tightly tied to their cellular compartments. Next, parallel analyses of the insoluble proteome revealed that distinct cellular compartments experience hampered turnover, in part due to misfolding. Finally, we discovered that age-associated fluctuations in the activity of the ubiquitin proteasome system were linked to the turnover of the catalytic core subunits. Taken together, our study provides a proteome-wide atlas of protein turnover across the aging continuum and highlights a link between the turnover of individual proteasome subunits and the age-associated decline in proteasome activity.

Disclosures: N.R. Rao: None. **A. Upadhyay:** None. **J.N. Savas:** None.

Poster

PSTR128. Aging-Related Molecular and Cellular Changes

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR128.05/D40

Topic: C.01. Brain Wellness and Aging

Support: NIH Grant F31AG074628

Title: Trem2-deficiency Hastens and Exacerbates Age-related Myelin Degeneration via Galectin-3

Authors: *T. MCCRAY, S. BISSEL, B. LAMB;
Stark Neurosciences Res. Inst., Indianapolis, IN

Abstract: Aging is the greatest known risk factor for various dementias and neurodegenerative diseases. Myelin degeneration often serves as an early pathological and clinical indicator of these diseases but is also a normal part of aging; albeit, to a lesser extent. Despite this, little is known about how age-related degeneration could contribute to neurodegenerative disease. Microglia participate in a variety of white matter events from demyelination to remyelination. The microglial innate immune receptor triggering receptor expressed on myeloid cells 2 (TREM2) modulates white matter pathology through activation and proliferation in response to demyelination, regulation of cholesterol metabolism during myelin phagocytosis, and release of trophic factors that aid in remyelination. The β-galactosidase-binding protein galectin-3 (gal-3), recently identified as a ligand for TREM2, also responds to white matter damage by acting as a crucial signal to initiate myelin phagocytosis, drive remyelination, and adapt to ER stress from accumulating lipid debris. We hypothesize that TREM2 prevents worsened age-related myelin degeneration through interactions mediated by gal-3. In this study, we investigate the effect of Trem2 on myelin degeneration in aged wildtype and Trem2-knockout mice. Utilizing a combination of immunohistochemistry, electron microscopy, and RNA/protein analyses we show Trem2-deficieny results in increased damage, altered signaling, and shifted cell populations in aged white matter. Further, to dissect the role of gal-3 in the response of TREM2, primary microglial cells were harvested from wild-type and Trem2-knockout mice to assess myelin phagocytosis in the presence of recombinant gal-3 or pharmacological gal-3 inhibitors. We show overlapping phagocytic deficits, lysosomal dysfunction, ER stress, and lipid droplet accumulation in microglia that were either Trem2-deficient or treated with gal-3 inhibitors. These data support a role for Trem2-dependent interactions with gal-3 in age-related myelin degeneration and suggest a basis for how dysfunctional microglia could contribute to disease pathology.

Disclosures: T. McCray: None. **S. Bissel:** None. **B. Lamb:** None.

Poster

PSTR128. Aging-Related Molecular and Cellular Changes

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR128.06/D41

Topic: C.01. Brain Wellness and Aging

Title: Long-term administration of resveratrol improves locomotor activity and declarative memory by reducing oxidative stress and brain cell loss in old rats.

Authors: *D. JUÁREZ SERRANO[1], I. CESAR ARTEAGA[1], S. TREVIÑO MORA[1], O. VERA LÓPEZ[2], A. NAVARRO CRUZ[2], G. FLORES[3], A. DIAZ[1];
[1]Posgrado en Ciencias Químicas, BQyBM, [2]Bioquímica-Alimentos, [3]Inst. de Fisiología, BENEMÉRITA UNIVERSIDAD AUTÓNOMA DE PUEBLA, PUEBLA, Mexico

Abstract: The cognitive functions of people over 60 years of age have been diminished, due to the structural and functional changes that the brain has during aging. Neuronal deterioration is different in each brain region. The greatest deterioration is reported in the frontal and temporal cortex, in addition to the hippocampus and cerebellum. In recent years, it has been shown that a therapeutic option for neurodegenerative processes and aging is antioxidant therapy. One of the most important polyphenols is resveratrol (trans-3,5,4-trihydroxystilbene) (RSVL). However, there are no reports of the use of RSVL administered from the juvenile stage and ending in the senile stage with the purpose of evaluating its effect on brain aging. The objective of this work was to evaluate the effect of RSVL on oxidative stress and cell loss in the frontal cortex, hippocampus, and cerebellum of 20-month-old rats and its consequences on recognition memory and motor behavior. Wistar male rats (n=24, 70 - 100 g of weight) of 30 days old were used, ramdomly divided into two groups: the control group (n=12) and the RSVL-treated group (n=12) with oral administration. RSVL was dissolved in a physiological saline solution (SS) as a vehicle at a dose of 10 mg/kg. The Novel object recognition task (NORt) was performed at the end of RSVL treatment (18 months). ROS quantification and lipid peroxidation was performed. In addition, the activity of superoxide dismutase (SOD), catalase (CAT) and the glutathione system comprising total GSH, reduced GSH, oxidized GSH (GSSG) and enzymes such as glutathione reductase (GR), glutathione S-transferase (GST) and glutathione peroxidase (GPx) were analyzed. Rats treated with RSVL showed an improvement in locomotor activity and in short- and long-term recognition memory. Likewise, the concentration of reactive oxygen species and lipid peroxidation decreased significantly in the group with RSVL, coupled with an improvement in the activity of the antioxidant system. Finally, with the help of hematoxylin and eosin staining, it was shown that chronic treatment with RSVL prevented cell loss in the brain regions studied. Our results demonstrate the antioxidant and neuroprotective capacity of RSVL when administered chronically. This strengthens the proposal that RSVL could be an important pharmacological option to reduce the incidence of neurodegenerative diseases that affect older adults.

Disclosures: D. Juárez Serrano: None. **I. Cesar Arteaga:** None. **S. Treviño Mora:** None. **O. Vera López:** None. **A. Navarro Cruz:** None. **G. Flores:** None. **A. Diaz:** None.

Poster

PSTR128. Aging-Related Molecular and Cellular Changes

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR128.07/D42

Topic: C.01. Brain Wellness and Aging

Support: NIH Grant AG066171

Title: Slow wave activity disruptions and memory impairments in a mouse model of aging

Authors: *L. YU[1,2], A. N. RUSS[1,2], M. ABEDIN[1,2], M. ALGAMAL[1,2], Q. ZHAO[1,2], M. R. MILLER[1,2], S. J. PERLE[1,2], K. V. KASTANENKA[1,2];
[1]Massachusetts Gen. Hosp., Boston, MA; [2]Harvard Med. Sch., Boston, MA

Abstract: The aging population suffers from memory impairments. Slow-wave activity (SWA) is composed of slow (0.5-1 Hz) and delta (1-4 Hz) oscillations, which play important roles in long-term memory and working memory function respectively. SWA disruptions might lead to memory disturbances often experienced by the elderly, but the neural bases underlying aberrant SWA are not understood. In this study, we conducted behavioral tests assessing working and long-term memory function in young (3 month old) and aged (24 month old) C57BL/6J mice, a widely recognized mouse model of aging. In vivo calcium imaging using multiphoton microscopy was used to monitor the activity of excitatory and inhibitory neurons in the cortices of these mice under isoflurane anesthesia. SWA was monitored using RH2080 voltage-sensitive dye (VSD) imaging through cranial windows. Aged mice exhibited impairments in working memory and fear acquisition, but not in fear recall dependent on memory consolidation during sleep. Impairments in neuronal function were observed with calcium imaging. Specifically, inhibitory neurons were hypoactive in aged mice. VSD imaging revealed aberrant synchronization of spontaneous neuronal activity in the cortices of aged mice. Notably, our observations revealed that aged mice exhibited no significant alterations in slow oscillations, whereas there was a significant increase in delta power when compared to young mice. Combined, these reported neural activity disruptions might underly memory impairments in aged C57BL/6J mice and could lead to identifying novel targets for therapeutic development aimed at improving memory function in the aging population.

Disclosures: L. Yu: None. **A.N. Russ:** None. **M. Abedin:** None. **M. Algamal:** None. **Q. Zhao:** None. **M.R. Miller:** None. **S.J. Perle:** None. **K.V. Kastanenka:** None.

Poster

PSTR128. Aging-Related Molecular and Cellular Changes

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR128.08/D43

Topic: C.01. Brain Wellness and Aging

Title: Single-cell transcriptomic atlas of the mammalian neurogenic niche reveals exercise as a countermeasure for brain aging

Authors: *W. XIANG[1], W. PAN[2], L. PENG[2], J. LEVI[1], A. IBRAYEVA[3], M. M. BAY[4], M. LIU[1], M. A. BONAGUIDI[5];

[1]Dept. of Stem Cell Biol. and Regenerative Med., [2]USC, [3]Broad CIRM Ctr. At USC, [4]Stem Cell & Regenerative Med., [5]Stem Cell Biol. & Regenerative Med., USC, Los Angeles, CA

Abstract: The hippocampus dentate gyrus serves as a microenvironment niche to support the self-renewal and differentiation of neural stem cells throughout life. Yet, the generation of neurons, glia, and overall hippocampus function deteriorates with age through unclear mechanisms. We have previously found neural stem cells undergo early cellular aging in the mature brain and that exercise slows this process. Here, we investigate molecular aging and exercise interventions on neurogenic niche cell types at the systems level. 7431 cells from the dentate gyrus of young and mature mice plus upon long-term running and short-term running were analyzed by mid-depth single-cell RNA sequencing. Systematic bioinformatics approaches identify mechanisms by which multiple niche cells exhibit early molecular aging and how exercise rescues these aging effects. Remarkably, other niche cells remain resistant to early aging, continue to undergo development in the adult brain and benefit from exercise through aging-independent mechanisms. We additionally interrogated the cross-talk interactions between neural stem cells and their niche components in young, aging, and exercise contexts to derive a comprehensive cell-cell communication network of the neurogenic niche. Our findings provide a valuable systems-level resource to explore early aging in the neurogenic niche, deconvolve complex intercellular communications, and reveal potential therapeutic targets of how exercise slows the aging process.

Disclosures: W. Xiang: None. **W. Pan:** None. **L. Peng:** None. **J. Levi:** None. **A. Ibrayeva:** None. **M.M. Bay:** None. **M. Liu:** None. **M.A. Bonaguidi:** None.

Poster

PSTR128. Aging-Related Molecular and Cellular Changes

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR128.09/D44

Topic: C.01. Brain Wellness and Aging

Support: NIA Grant AG060778

Title: Impact of age on cholinergic effector systems underlying persistent firing of principal neurons in the rat basolateral amygdala

Authors: *T. SAHAGIAN, J. THINSCHMIDT, S. HARDEN, J. BIZON, B. SETLOW, C. J. FRAZIER;
Pharmacodynamics, Univ. of Florida, Gainesville, FL

Abstract: Persistent firing is a term often used to describe long-lasting neuronal firing that occurs in response to a transient stimulus. This type of neuronal behavior has previously been implicated in a variety of working memory and temporal association tasks. We evaluated the ability of an acetylcholine receptor (AChR) agonist, carbachol, to promote persistent firing in principal neurons of the basolateral amygdala (BLA PN), a brain region that receives one of the

densest cholinergic projections in the CNS. BLA PNs appear to play an important modulatory role in a variety of behaviors, including cost/benefit decision-making. Interestingly, recent work has highlighted that aging alters the contributions of BLA PNs to temporally specific aspects of choice behavior. In the current study, we used whole-cell patch clamp recordings in acute tissue slices containing the BLA to carefully evaluate effector systems in BLA PNs that are coupled to activation of AChRs. We identified carbachol mediated and M1 receptor dependent modulation of multiple effectors that collectively depolarize BLA PNs, reduce activity-induced inhibitory responses, and promote a long lasting, often suprathreshold afterdepolarization. Additional data indicate that the cholinergic afterdepolarization depends heavily on activation of a calcium-activated non-selective cation channel (I_{CAN}), and directly promotes persistent firing. Preliminary data in adult vs. aged animals further indicate that, in the presence of an AChR agonist, aging alters activity-dependent activation of I_{CAN} in a manner that is likely to impact persistent firing.

Disclosures: **T. Sahagian:** None. **J. Thinschmidt:** None. **S. Harden:** None. **J. Bizon:** None. **B. Setlow:** None. **C.J. Frazier:** None.

Poster

PSTR128. Aging-Related Molecular and Cellular Changes

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR128.10/D45

Topic: C.01. Brain Wellness and Aging

Support: CONAHCyT Grant 847273 to LAH
CONAHCyT Grant 252808 to GF

Title: Erythropoietin Attenuates Age-Induced Neuronal Plasticity and Cognitive Declines in the C57BL6 Mice

Authors: *L. AGUILAR HERNÁNDEZ[1,2,3], R. ALEJANDRE[2], J. MORALES-MEDINA[5], A. DÍAZ[4], S. TREVIÑO[4], G. FLORES[3];
[1]Neuropsychiatry laboratory, Inst. Politécnico Nacional, Puebla, Mexico; [2]Escuela Nacional de Ciencias Biológicas, Inst. Politécnico Nacional, México, Mexico; [3]Inst. de Fisiología, [4]Facultad de Ciencias Químicas, Benemérita Univ. Autónoma de Puebla, Puebla, Mexico; [5]Ctr. de Investigación en Reproducción Animal, CINVESTAV-Universidad Autónoma de Tlaxcala, Tlaxcala, Mexico

Abstract: Aging is a physiological process. The brain is one of the organs most affected by aging, observing a lower capacity to maintain the integrity of neural networks, and a reduction in neuronal plasticity, resulting in a less efficient cognitive function. It manifests itself functionally primarily as losses in memory and learning. To allow the greatest efficiency in neuronal plasticity and reduce the cognitive deterioration typical of the elderly population, it is necessary to implement substances that provide protection, repair, and maintenance to brain tissue. Erythropoietin (EPO) is an endogenous human protein hormone whose main function is the

stimulation of erythropoiesis. EPO is produced in response to plasmatic hypoxia and subsequently interacts with its specific receptors on erythroid progenitor cells. In these cells, it triggers proliferation, differentiation, and anti-apoptosis effects that stimulate their maturation into functional erythrocytes. It has been shown that neurons and glial cells have EPO receptors and the ability to produce this hormone. At the brain tissue, EPO triggers effects such as neuronal protection and repair.In the present work, the effect of chronic EPO treatment was assessed in mice of 3, 6, and 12 months of age. The novel object recognition test determined if EPO or age caused alterations in recognition memory. The density and types of dendritic spines were evaluated using the Golgi-cox technique. Neuronal density was quantified by stereology technique, and the presence of BDNF, synaptophysin, C-Fos, GFAP, and Iba1 was estimated by immunohistochemistry. The regions analyzed were prefrontal cortex, hippocampus, and amygdala. In addition, the modifications in blood count induced by EPO were analyzed. Our results show an improvement in neuronal plastic capacities that slow down age-induced memory decline. A structural improvement in the dendritic spines was observed, accompanied by a reduction in neuroinflammation. There is also a recovery in the expression of synaptic proteins that could be correlated with the improvement in the morphology of dendritic spines.

Disclosures: **L. Aguilar Hernández:** None. **R. Alejandre:** None. **J. Morales-Medina:** None. **A. Díaz:** None. **S. Treviño:** None. **G. Flores:** None.

Poster

PSTR128. Aging-Related Molecular and Cellular Changes

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR128.11/D46

Topic: C.01. Brain Wellness and Aging

Support: University of Alabama Pilot Project Program

Title: Neuroprotective Potential of Fucoxanthin in Middle Aged Sprague-Dawley Rats

Authors: *H.-A. PARK, K. FERDOUS, L. CIESLA, R. N. CORRELL, A. ELLIS;
Univ. of Alabama, Tuscaloosa, AL

Abstract: Fucoxanthin is a marine carotenoid found in brown seaweed. Fucoxanthin exhibits strong antioxidant properties and is shown to produce neuroprotective properties against pathological processes in the brain. Although the kinetics of fucoxanthin are well studied in various organs including the liver, adipose tissues, and blood, availability of fucoxanthin in the brain has not been researched. In this study hypothesized that orally administrated fucoxanthin is delivered to and directly protects the brain. Additionally, we screened genes that are sensitive to fucoxanthin supplementation. Middle-aged (18-month old) male Sprague-Dawley rats were orally gavaged with fucoxanthin (1mg/kg) or vehicle oil for 4 weeks. After supplementation, animals were euthanized, followed by transcardial perfusion, and brain tissues were collected and analyzed. Fucoxanthin supplemented animals showed greater levels of fucoxanthin and

fucoxanthionol in the brain compared to the control group. Additionally, whole brain tissues were analyzed using the rat LncRNA microarray. Oral supplementation of fucoxanthin upregulated 2666 genes and downregulated 2936 genes. Oral supplementation of fucoxanthin changed genes involved in cytokine-cytokine receptor interaction, inflammatory mediator regulation, and calcium signaling pathway. This study shows that fucoxanthin is found in the brain after oral administration to regulate gene expression.

Disclosures: **H. Park:** None. **K. Ferdous:** None. **L. Ciesla:** None. **R.N. Correll:** None. **A. Ellis:** None.

Poster

PSTR128. Aging-Related Molecular and Cellular Changes

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR128.12/D47

Topic: C.01. Brain Wellness and Aging

Support: NIH R01AG065206 (CJS)
NIH RF AG080742 (KIA)
NIH RF AG070839-01 (KIA)
Fondation Bertarelli (JH)
NIH T32MH020016 (JH)

Title: A subset of hippocampal genes is restored to youthful levels in aged mice by inhibiting the prostaglandin E2 receptor

Authors: *J. HUANG, C. J. SHATZ, K. I. ANDREASSON;
Stanford Univ., Stanford, CA

Abstract: Aberrant systemic immune responses are a shared feature across neurological conditions associated with impaired cognitive function such as aging, disease, and injury. Targeting the immune response may be a key to resolving cognitive deficits. Remarkably, the Andreasson lab has found that dampening proinflammatory prostaglandin E2 receptor (EP2) signaling in immune cells restores learning and memory in aged mice. Two-week treatment with a brain-penetrant EP2 inhibitor (C52) is sufficient to restore youthful hippocampal LTP and learning/memory in aged mice. However, it is still unknown how this intervention, which acts on immune cells, restores function to neurons and ultimately leads to improved learning and memory. Initial studies have discovered that microglia are restored to homeostatic glucose metabolism and morphology with EP2 inhibition, but these have only scratched the surface of how C52 rejuvenates LTP and cognition. We hypothesized that treating the maladaptive aging immune system with C52 reverses the aged hippocampus to a youthful gene expression profile. To test this hypothesis, we performed bulk RNA-sequencing using hippocampi from young and aged C57Bl/6 mice treated with either a vehicle or C52 solution for two weeks. Differential gene expression analysis and pathway enrichment in the aged vehicle compared to the young vehicle

showed an upregulation of well documented age-associated pathways including complement cascade, immune response, and regulation of antigen processing and presentation. Notably, C52 treatment in aged mice restores expression of a subset of age-associated genes to similar levels as those in the young vehicle condition. Restored pathways include GPCR downstream signaling and metal ion transport. Validation of candidate genes is currently ongoing. Together, this work shows that it is possible to restore age-related changes in hippocampal gene expression to more youthful expression signatures by dampening proinflammatory EP2 signaling, adding to previous observations of the effect of EP2 inhibition on microglial metabolism and morphology, and opening the door to identifying molecular candidates for further study.

Disclosures: J. Huang: None. **C.J. Shatz:** None. **K.I. Andreasson:** None.

Poster

PSTR128. Aging-Related Molecular and Cellular Changes

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR128.13/D48

Topic: C.01. Brain Wellness and Aging

Support: James S. McDonnell Foundation (220020346) to Paul S. Garcia

Title: The effects of elimination of the alpha-4 subunit of GABA-A receptors on general anesthesia and recovery biomarkers in aged mice

Authors: *M. MANSOURI[1], M. GRAVES[1], C. TROYAS[2], T. Z. CASSIM[1], P. S. GARCÍA[1]; [1]Anesthesiol., Columbia Univ., New York, NY; [2]Anesthesiol. and Intensive Care Med., Tech. Univ. of Munich, Munich, Germany

Abstract: Introduction: Recovery from general anesthesia (GA) involves restoration of cortical communication to cortical and subcortical areas. Clinical studies suggest that aging may impair recovery from GA. We have previously shown that deletion of *gabra4* gene, which encodes alpha-4 subunit containing GABA-A receptors (α4-GABA) expression in the thalamus, accelerates emergence from isoflurane in mice. This study aimed to investigate whether recovery from isoflurane and propofol GA is impaired in aged α4-GABA knockout (KO) mice. **Methods:** Both isoflurane and propofol were used to test anesthetic sensitivity, emergence (return of righting reflex; RORR), and recovery (sticky dot notice) following GA in young adult (3-4 months) and middle-aged (11-13 months) α4-GABA KO and wild-type (WT) mice. Spectral analysis and time intervals of suppressed cortical activity were performed during anesthesia. Cognitive performance was also evaluated using spontaneous Y-maze test. **Results:** Minimal alveolar concentration to suppress movement by isoflurane (MAC immobility) was not changed in both young and middle-aged KO mice as compared to the WT littermates. Further, although lack of α4-GABA in young mice can hasten emergence (decrease time to RORR), delays in RORR were significantly increased in both WT and KO middle-aged mice under either isoflurane or propofol anesthesia. Additionally, aging delayed recovery after GA in both

treatment groups defined by sticky-dot notice regardless of the genotype. Pharmacologic studies with gaboxadol in isoflurane-treated groups showed that time to RORR was significantly increased in both young and middle-aged WT mice but had minimal effects on KO mice. Further, gaboxadol delayed time to sticky dot notice in both young and middle-aged mice regardless of the genotype. The young α4-GABA KO mice exhibited EEG indicative of more burst suppression than the WT mice during propofol and low-dose isoflurane (1.5%) anesthesia. An increase in the isoflurane dose (2%) was associated with an increase in the BSR for both the WT and KO mice. These events were altered with age. Finally, genetic deletion of α4-GABA had no effects on the Y-maze correct alternations even after both isoflurane and propofol anesthesia. **Conclusion:** These findings suggest that the *in vivo* effects of isoflurane and propofol on KO mice depend on age. Further, we confirm age-associated delays in recovery from GA. We tentatively conclude that impairment of cortical signaling involving the thalamus is involved in producing these results and speculate that age-related changes in excitatory-inhibitory balance modify the effects of elimination of the α4-subunit of the GABA-A receptor.

Disclosures: **M. Mansouri:** None. **M. Graves:** None. **C. Troyas:** None. **T.Z. Cassim:** None. **P.S. García:** None.

Poster

PSTR128. Aging-Related Molecular and Cellular Changes

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR128.14/D49

Topic: C.01. Brain Wellness and Aging

Support: Alzheimer's Association Research Fellowship Diversity AARFD-21-853104
BrightFocus Foundation A2022048S
Goizueta Alzheimer’s Disease Research Center P30 AG066511
National Institutes of Health NINDS R01-NS-109226
Packard Award in Science and Engineering

Title: Non-invasive flicker neurostimulation boosts resilience to psychological stress

Authors: *T. FRANKLIN[1], M. GOODSON[2], T. GAJELLI[3], A. PRICHARD[1], C. RUTLEDGE[3], A. SINGER[1];
[1]Biomed. Engin., Georgia Inst. of Technology- Emory Univ., Atlanta, GA; [2]Biomed. Engin., [3]Georgia Inst. of Technol., Atlanta, GA

Abstract: Chronic stress promotes life-long risk for neuropsychiatric decline by disrupting inflammatory signaling, and compromising synaptic health and plasticity. Our lab and others have demonstrated that non-invasive gamma sensory flicker neurostimulation modulates inflammatory signaling, restores microglia function, and improves cognitive performance in mouse models of amyloidosis. However, the effects of sensory flicker in the context of stress are

unknown. Accordingly, our goal for this study was to determine how sensory flicker stimulation mitigates neuropsychiatric-like behavioral deficits and rescues glia and synapse pathology following chronic stress. We hypothesized that non-invasive sensory flicker would boost successful behavioral adaptability, or resilience, to chronic psychological stress in a frequency specific manner by mitigating stress-induced synaptic remodeling. Daily audiovisual flicker intervention at multiple frequencies was introduced concomitantly with daily stress exposure (28 days) in male and female C57BL6 and Thy1-GFP mice. The mice were then tested for stress-induced anxiety-like behavior and anhedonia using a range of behavioral tests. Because synaptic remodeling and microglia reactivity are thought to underlie stress-induced anxiety and anhedonia, we quantified spine density changes in Thy1-GFP mice and quantified IBA1-labeled microglia morphology change, a reliable proxy for microglia reactivity, in the medial prefrontal cortex (mPFC) using semi-automated Imaris imaging software. We show for the first time that flicker stimulation results in behavioral resilience in stressed mice in a sex-, and frequency-specific manner that coincides with modulation of stress-induced microglia morphological changes in the mPFC. Our findings indicate that audio-visual flicker protects against stress-induced behavioral deficits and modulates glia-neuron interaction in a sex- and frequency-specific manner. Together, these findings show flicker intervention, tuned by frequency, improves stress pathology and may prevent stress-induced neuropsychiatric health decline in conditions with sex dimorphic symptoms and prevalence.

Disclosures: **T. Franklin:** None. **M. Goodson:** None. **T. Gajelli:** None. **A. Prichard:** None. **C. Rutledge:** None. **A. Singer:** E. Ownership Interest (stock, stock options, royalty, receipt of intellectual property rights/patent holder, excluding diversified mutual funds); Cognito.

Poster

PSTR128. Aging-Related Molecular and Cellular Changes

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR128.15/D50

Topic: C.01. Brain Wellness and Aging

Support: NIH MH120734-01
NIH U24 AG072701-02S1

Title: Age-related fatigue worsens brain aging effects on cognition in older adults with neurodegeneration

Authors: *M. ANTHONY[1], A. TURNBULL[2], F. LIN[2], D. TADIN[1];
[1]Univ. of Rochester, Rochester, NY; [2]Stanford Univ., Stanford, CA

Abstract: Fatigability is a common age-related phenomenon in which perceived mental and/or physical exertion interferes with everyday activities. Increased fatigue is associated with negative functional outcomes in older adults, including worse cognitive impairment. Older adults with more severe neurodegeneration also perceive greater fatigue and exhibit less efficient brain

energy expenditure compared to healthy older adults. Fatigue may be a behavioral indicator of brain aging; however, supporting empirical work is limited. To this end, we performed moderation analyses to examine the association between fatigue (Multidimensional Fatigue Inventory), and cognition (episodic memory or executive function) in older adults with and without neurodegeneration (Alzheimer's disease-signature cortical thickness). Results showed that fatigue was significantly related to cognition ($t = -2.55$, $p = 0.012$), and this effect was significantly stronger for older adults with worse neurodegeneration (*interaction term:* $t = 2.50$, $p = 0.014$). We validated our findings using data from a double-blinded intervention (primarily aimed at understanding brain stimulation effects on mood) to examine the mechanistic relationships between fatigue (Visual Analogue Scale), neurodegeneration, and cognitive plasticity (change in episodic memory or executive function, between baseline and post-intervention) in older adults with neurodegeneration. Fatigue was assessed at 14 timepoints over one month, with level of fatigue and stability of energy defined as mean and SD, respectively. Fatigue was related to cognition ($t = 2.30$, $p = 0.018$), and this effect was significantly stronger for older adults with worse neurodegeneration (*interaction term:* $t = -0.76$, $p = 0.029$). Stability of energy was related to cognition ($t = -4.83.$, $p = 0.01$), and this effect was significantly stronger for older adults with worse neurodegeneration (*interaction ter*m: $t = 1.63$, $p = 0.016$). Fatigue was also related to executive function ($t = 1.80$, $p = 0.031$); the effect was significantly stronger for older adults with worse neurodegeneration (*interaction term:* $t = -0.65$, $p = 0.032$). Stability of energy was related to executive function (*interaction term:* $t = -3.69$, $p = 0.019$); the effect was significantly stronger for older adults with worse neurodegeneration (*interaction ter*m: $t = 1.32$, $p = 0.017$). Findings provide evidence that fatigue explains why some older adults show better cognitive function or more cognitive improvement than others, regardless of neurodegeneration, with implications for understanding how brain aging-relevant factors influence the efficacy of cognitive interventions in older adults.

Disclosures: **M. Anthony:** None. **A. Turnbull:** None. **F. Lin:** None. **D. Tadin:** None.

Poster

PSTR128. Aging-Related Molecular and Cellular Changes

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR128.16/D51

Topic: C.01. Brain Wellness and Aging

Support: HABS-HD U19AG078109

Title: Depression medication and hippocampal subfield volume in older adults

Authors: *A. M. TWISSELMANN[1], V. R. TENNANT[1], B. J. HALL[1], A. MAHAJAN[1], A. KIM[1], M. L. DAVISON[1], K. WHEELER[1], C. R. K. CHING[1], *A. M. TWISSELMANN[2], J. HALL[5], M. T. BORZAGE[3], L. JOHNSON[5], A. W. TOGA[4], S. E. O'BRYANT[5], K. YAFFE[6], M. N. BRASKIE[1];
[1]Imaging Genet. Ctr., USC, Marina del Rey, CA; [2]Inst. for Neuroimaging, USC, Westminster,

CA; [3]Children's Hosp. Los Angeles, USC, Los Angeles, CA; [4]Inst. for Neuroimaging, USC, Marina del Rey, CA; [5]Univ. of North Texas, Fort Worth, TX; [6]Univ. of California at San Francisco, San Francisco, CA

Abstract: Major Depressive Disorder (MDD) and Alzheimer's Disease are highly comorbid conditions associated with smaller hippocampal volume and decreased memory performance. MDD is treatable with antidepressants, though there is conflicting literature on their impact on hippocampal volume, particularly subfield volume. We investigated the relationship between MDD medication status and hippocampal subfield volume in older adults. We examined 493 cognitively intact older adults with MDD from the Health and Aging Brain Study - Health Disparities with self-reported use of a medication primarily used to treat MDD and available hippocampal high resolution MRI scans (Siemens Skyra or Vida-3T) (Table 1). The scans were segmented into left and right averaged CA1, a region that included CA2, 3, and dentate gyrus (CA23DG), and subiculum using Automatic Segmentation of Hippocampal Subfields (ASHS) software. We used multiple linear regressions to investigate the relationships between treatment and CA1 and CA23DG volumes, which were previously negatively associated with MDD in this cohort. We covaried for age, gender, years of education, number of coronal MRI slices segmented, intracranial volume, and MRI scanner . A follow-up Welch's t-test evaluated the relationship between score on the geriatric depression scale (GDS) and medication status. MDD medication was associated with smaller CA1 (β;;= -0.13, p=0.03), with a trend-level relationship in the CA23DG (β;;= -0.12, p=0.09). Those on MDD medications had a lower average score on the GDS than those who were not medicated (t = 2.63, p=0.01), suggesting the effectiveness of medication on depressive symptoms. One possible explanation for the relationship between CA1 volume and MDD medication status is that those who are on medication had more severe MDD pre-medication, aligning with the findings of the largest study of MDD (n=8927), which may have had deleterious effects on brain aging. Further investigation is needed to explore the impact of MDD severity and medication status on hippocampal subfield volume.

	Medicated	Non-Medicated
N	211	282
Sex (n)	42M/169F	66M/216F
Ethnicity (n) *	123NHW/55MA/33AA	95NHW/136MA/51AA
Mean age (years)	64.06 ± 7.54	64.15 ± 8.37
Mean education (years) *	13.96 ± 3.87	12.07 ± 4.92
Score on GDS (points) *	8.92 ± 6.68	10.49 ± 6.43

Table 1. The demographic breakdown of the sample population. Chi-squared tests were performed on sex and ethnicity subsets and t-tests were performed on age, education, and GDS score. Significant relationships ($p<0.05$) are marked with *. NHW=non-Hispanic white, MA=Mexican American , AA=African American.

Disclosures: **A.M. Twisselmann:** None. **V.R. Tennant:** None. **B.J. Hall:** None. **A. Mahajan:** None. **A. Kim:** None. **M.L. Davison:** None. **K. Wheeler:** None. **C.R.K. Ching:** None. **A.M. Twisselmann:** None. **J. Hall:** None. **M.T. Borzage:** None. **L. Johnson:** None. **A.W. Toga:** None. **S.E. O'Bryant:** None. **K. Yaffe:** None. **M.N. Braskie:** None.

Poster

PSTR128. Aging-Related Molecular and Cellular Changes

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR128.17/D52

Topic: C.01. Brain Wellness and Aging

Support: VA Puget Sound Healthcare System Grant: GRANT13570727

Title: Chronic adrenergic blockade enhances glymphatic pathway function in a murine model

Authors: M. SEVAO[1], *J. CHO[2];
[1]VA Puget Sound/University of Washington, Seattle, WA; [2]Univ. of Washington, Seattle, WA

Abstract: Chronic adrenergic blockade enhances glymphatic pathway function in a murine model. Mathew Sevao, Joshua Cho, Murray Raskind, Jeffrey IliffThe glymphatic system has been characterized as the brain's waste clearance system, utilizing a series of perivascular channels to exchange cerebrospinal fluid (CSF) and interstitial fluid and facilitate clearance of interstitial solutes such as amyloid β and tau. Previous studies in rodents and humans have demonstrated that glymphatic function is enhanced during sleep and is suppressed during wakefulness. Glymphatic function is impaired in animal models of aging, cerebrovascular dysfunction, and traumatic brain injury, each of which has been identified clinically as non-genetic risk factors for neurodegenerative conditions such as Alzheimer's disease, Parkinson's disease, and chronic traumatic encephalopathy. These findings suggest that glymphatic impairment may contribute to the development of age-related and post-traumatic neurodegenerative conditions, and that therapeutic approaches that enhance glymphatic function could be utilized to treat or prevent these neurodegenerative conditions. Prazosin, an alpha-1 adrenergic receptor antagonist, is used clinically in the setting of trauma nightmares, where it has been shown to improve sleep disturbances. Little is known about its impact on the glymphatic system, however. Here, we hypothesized that glymphatic function is enhanced by blockade of central adrenergic tone. Using a murine model, we measured glymphatic function following a 7-day treatment of prazosin administered in drinking water at 45mg/L. Glymphatic function was quantified using an intracisternal co-injection of infrared (IR) and conventional fixable fluorescent tracer. CSF tracer distribution was evaluated through a newly validated in vivo dynamic imaging technique over a course of 42 minutes, and paired with whole-slice fluorescent imaging, allowing a histological assessment of glymphatic functioning. We found a significant enhancement of glymphatic function in the prazosin treated group compared to the control ($p<0.05$ for 32-38min, $p<0.01$ for 40-42min, $n=8$ per group). We further evaluated sex differences using ex vivo whole brain imaging and found the males to have a greater

enhancement compared to the females (1.013 +/- 0.1530, p=0.0165, n=8 per group). These findings may provide further insight on the underlying mechanisms that promote neurodegeneration and may be studied further to provide improved treatments for patients with neuropathological conditions linked to glymphatic dysfunction.

Disclosures: **M. Sevao:** None. **J. Cho:** None.

Poster

PSTR129. Alzheimer's Disease: Molecular and Cellular Mechanisms I

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR129.01/D53

Topic: C.02. Alzheimer's Disease and Other Dementias

Title: Acitretin, an upregulator of alpha-secretase (ADAM10), strengthens hippocampal long-term potentiation in Aβ-treated slices and inthe 5xFAD mouse model of Alzheimer’s Disease

Authors: *K. D. PARFITT, A. H. SHERIDAN, R. S. SIVAKUMAR;
Neurosci., Pomona Col., Claremont, CA

Abstract: Secreted APP alpha (sAPPα) is a product of non-amyloidogenic APP processing that has neurotrophic and neuroprotective properties and enhances hippocampal long term potentiation (LTP) *in vivo* and *in vitro*. In addition, over-expression of sAPPα can prevent deficits in spatial memory seen in the APP/PS1 mouse model of Alzheimer’s Disease. Recently, acitretin, an FDA-approved synthetic retinoid, has been demonstrated to increase the concentration of sAPPα in an α-secretase (ADAM-10)-dependent manner. In addition, nine-day intraperitoneal (i.p.) treatment with acitretin reverses behavioral deficits in 4-month-old 5XFAD mice. In the present study, we examined the impact of acitretin treatment on hippocampal long-term potentiation (LTP) *in vitro* in both C57BL/6 and 5xFAD mice by extracellularly recording EPSPs at CA3/CA1 Schaffer collateral synapses. We predicted that acitretin treatment would raise CNS sAPPα levels and thereby enhance LTP in slices prepared from treated mice. We found that acitretin treatment of C57BL/6 mice (five days of treatment; 10 mg/kg i.p.) can produce LTP induced by an otherwise sub-threshold half-theta burst stimulation (half-TBS). Application of a half-TBS (5 trains (5 Hz) of 5 pulses (100 Hz)) induced post-tetanic potentiation (PTP), but not LTP, in slices from mice treated with vehicle. In contrast, slices from mice treated with acitretin showed significantly enhanced PTP and stable LTP (148.16 ± 19.51% of baseline, vs 100.73 ± 2.85% of baseline in vehicle-treated mice; $p<0.01$). The 5-day acitretin treatment also prevented the deficits in LTP seen following treatment of slices with $A\beta_{25-35}$ (200nM), an active fragment of β-amyloid. LTP induced by two full TBS (10 trains (5 Hz) of 5 pulses (100 Hz); 148.04 ± 11.89% of baseline) was significantly reduced in slices treated with $A\beta_{25-35}$ (119.7 ± 9.8 of baseline), but these deficits were prevented by the pretreatment of mice with acitretin (177.57 ± 42.99% of baseline). In a parallel investigation in 8-month-old 5xFAD mice, we also observed that deficits in LTP were ameliorated by acitretin treatment ($p < 0.05$, 5xFAD: LTP = 127.05 ± 15.03% of baseline; 5xFAD + Acitretin: LTP = 167.39 ± 10.50%). Taken together,

these results offer preliminary evidence supporting the utility of acitretin as a novel therapeutic target for AD, paving the way for possible drug repurposing of retinoids for AD treatment.

Disclosures: **K.D. Parfitt:** None. **A.H. Sheridan:** None. **R.S. Sivakumar:** None.

Poster

PSTR129. Alzheimer's Disease: Molecular and Cellular Mechanisms I

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR129.02/D54

Topic: C.02. Alzheimer's Disease and Other Dementias

Support: NIH Grant R01AI132414-04
Philanthropic gift by The Stead Family Foundation

Title: Evaluating the Influence of HCMV Infection on Alzheimer's Disease Pathology

Authors: ***J. W. ADELMAN**[1], S. ROSAS-ROGERS[2], S. S. TERHUNE[2], A. D. EBERT[1]; [1]Cell Biology, Neurobio. and Anat., [2]Microbiology and Immunol., Med. Col. of Wisconsin, Milwaukee, WI

Abstract: Alzheimer’s Disease (AD) is a common, incurable neurodegenerative condition characterized by progressive memory deficits, behavioral aberrancies, and death. While its underlying mechanisms remain unclear, previous studies have highlighted roles for aberrant protein aggregation (e.g., Amyloid beta, phospho-Tau) and synaptic dysfunction in AD pathophysiology. Further, a growing body of research shows viral contribution to disease progression, particularly among human herpesviruses (HHVs). Specifically, Human Cytomegalovirus (HCMV, HHV5) has been shown to be associated with various AD phenotypes, including altered calcium signaling, increased amyloid beta (Aß) accumulation, and altered synaptic structure. Considering this, we hypothesized that HCMV infection potentiates AD-like phenotypes in neuronal populations. Using induced pluripotent stem cells (iPSCs) derived from healthy and AD-affected individuals, we generated a forebrain-specific 2D neuronal culture system to directly assess any impacts of HCMV infection (clinical strain TB40/E) on AD pathology in human neurons. Our preliminary data highlight a potential role of HCMV in limiting neurotransmission via downregulation of the synaptic machinery necessary for vesicle fusion (e.g., Syt1, Syp, SNAP25, Munc18-1, VAMP2). Of note, several of these targets are also dysregulated in AD and are potential biomarkers for disease progression. We postulate that HCMV reduces exocytosis rates via protein downregulation. Using a combination of biochemical techniques and live-cell imaging, we will assess HCMV’s effects on both individual synaptic proteins and the process of vesicle trafficking/fusion in both AD- and control-derived cells. Additionally, we have found that HCMV-infected neurons exhibit a slight increase in both intracellular Aß1-42 and p-Tau, whereas ELISAs examining neuron-conditioned media revealed HCMV-dependent reductions in extracellular Aß42/40 ratios. We hypothesize that HCMV alters the solubility of Aβ and Tau thereby reducing detection of soluble fractions as

we observe increases in insoluble protein accumulation. Finally, we will test whether blocking viral entry and/or inhibiting viral polymerase mitigates decreases in synaptic vesicle function and protein insolubility. Together, these data will elucidate the pathological impact of HCMV infection in human cortical neurons and identify potential targets of therapeutic intervention.

Disclosures: J.W. Adelman: None. **S. Rosas-Rogers:** None. **S.S. Terhune:** None. **A.D. Ebert:** None.

Poster

PSTR129. Alzheimer's Disease: Molecular and Cellular Mechanisms I

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR129.03/D55

Topic: C.02. Alzheimer's Disease and Other Dementias

Support: NIH Grant R01 NS117372
NIH Grant R21 NS121284
Simons Foundation Autism Research Initiative (SFARI) BTI award 551354
Brain and Behavior Research Foundation Young investigator award 27792

Title: Dysregulated Glial Genes in Alzheimer’s Disease Are Essential for Homeostatic Plasticity: Evidence from Integrative Epigenetic and Single Cell Analyses

Authors: *T. CUI[1,2], **Y. CAI**[2], P. YIN[2], P. PAGANELLI[2], S. VICINI[2], T. WANG[2]; [2]Pharmacol. and Physiol., [1]Georgetown Univ. Med. Ctr., Washington, DC

Abstract: Synaptic homeostatic plasticity is a fundamental form of regulation that stabilizes synaptic and neural physiology in the nervous system. Impairment of homeostatic regulation has been linked to synapse destabilization during the progression of Alzheimer’s Disease (AD). Recent advances in epigenetic and transcriptomic characterizations of the nervous system during aging and in AD have provided valuable insights into the molecular etiology of neurodegeneration. However, how abnormal epigenetic and transcriptomic alterations in different cell types in AD affect synaptic homeostatic plasticity remains to be elucidated. Different glial cell types play critical roles in controlling synaptic function during aging and in neurodegenerative disorders. Here, we investigated how the glial dysregulation of histone acetylation and transcriptomes in AD impacts synaptic homeostatic plasticity through computational analysis and electrophysiology-based genetic screens in *Drosophila*. Through integrative analysis of scRNA-seq and ChIP-seq data in the same cohort of AD patients, we identified cell type-specific signature genes that are transcriptionally dysregulated by histone acetylation. We systematically studied the function of the glial signature genes in regulating Presynaptic Homeostatic Potentiation in *Drosophila*. Remarkably, all the dysregulated glial-specific genes identified from the integrative computational analysis are necessary for controlling synaptic homeostatic plasticity in *Drosophila*. Thus, we provided genetic evidence

demonstrating that abnormal glial transcriptomic alterations in AD are closely associated with impairment of homeostatic plasticity in the nervous system. Overall, our computational and genetic analyses prioritize glial gene candidates that are potentially linked to homeostatic dysregulation in AD.

Disclosures: **T. Cui:** None. **Y. Cai:** None. **P. Yin:** None. **P. Paganelli:** None. **S. Vicini:** None. **T. Wang:** None.

Poster

PSTR129. Alzheimer's Disease: Molecular and Cellular Mechanisms I

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR129.04/D56

Topic:

Support: NINDS T32 NS061788
NIA AG054719
NIA AG063755
NIA AG068024
R01AG061800
U01AG061357
P30AG10161
P30AG72975
R01AG15819
R01AG17917
U01AG46152
U01AG61356

Title: Regional brain co-expression network analysis identifies NRN1 as a mediator of cognitive resilience to Alzheimer's disease

Authors: *D. PUGH[1], C. HURST[2], M. ABREHA[2], D. DUONG[2], E. DAMMER[2], D. A. BENNETT[3], N. SEYFRIED[2], J. HERSKOWITZ[1];
[1]UAB Neurosci. Grad. Programs, Birmingham, AL; [2]Emory Univ., Emory Univ., Atlanta, GA; [3]Rush Univ. Med. Ctr., Rush Univ. Med. Ctr., Chicago, IL

Abstract: The molecular mechanisms and pathways enabling certain individuals to remain cognitively normal despite high levels of Alzheimer’s disease (AD) pathology remain incompletely understood. These cognitively normal people with AD pathology are described as preclinical or asymptomatic AD (AsymAD) and appear to exhibit cognitive resilience to the clinical manifestations of AD dementia. Here we present a comprehensive network-based approach from cases clinically and pathologically defined as asymptomatic AD to map resilience-associated pathways and extend mechanistic validation. Multiplex tandem mass tag mass spectrometry (TMT-MS) proteomic data (n=7,787 proteins) was generated on brain tissue

from Brodmann area 6 and Brodmann area 37 (n=109 cases, n=218 total samples) and evaluated by consensus weighted gene correlation network analysis. Notably, neuritin (NRN1), a neurotrophic factor previously linked to cognitive resilience, was identified as a hub protein in a module associated with synaptic biology. To validate the function of NRN1 with regard to neurobiology to AD, we conducted microscopy and physiology experiments in a cellular model of AD. NRN1 provided dendritic spine resilience against amyloid-β (Aβ) and blocked Aβ-induced neuronal hyperexcitability in cultured neurons. To better understand the molecular mechanisms of resilience to Aβ provided by NRN1, we assessed how exogenous NRN1 alters the proteome by TMT-MS (n=8,238 proteins) of cultured neurons and integrated the results with the AD brain network. This revealed over-lapping synapse-related biology that linked NRN1-induced changes in cultured neurons with human pathways associated with cognitive resilience. Collectively, this highlights the utility of integrating the proteome from human brain and model systems to advance our understanding of resilience-promoting mechanisms and prioritize therapeutic targets that mediate resilience to AD.

Disclosures: **D. Pugh:** None. **C. Hurst:** None. **M. Abreha:** None. **D. Duong:** None. **E. Dammer:** None. **D.A. Bennett:** None. **N. Seyfried:** None. **J. Herskowitz:** None.

Poster

PSTR129. Alzheimer's Disease: Molecular and Cellular Mechanisms I

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR129.05/D57

Topic: C.02. Alzheimer's Disease and Other Dementias

Support: NIA AG067635
NIA AG061800
NIA AG054719
NIA AG063755
NIA AG068024
NIA AG061357
NINDS NS061788
NINDS NS095775

Title: Cross-platform synaptic network analysis of human entorhinal cortex identifies TWF2 as a modulator of dendritic spine length

Authors: *K. M. GREATHOUSE[1], C. K. WALKER[2], A. J. WEBER[3], N. T. SEYFRIED[5], J. H. HERSKOWITZ[4];
[1]Neurol., UAB, Birmingham, AL; [2]Neurol., Univ. of Alabama Birmingham, Birmingham, AL; [3]Neurol., [4]Univ. of Alabama at Birmingham, Birmingham, AL; [5]Emory Univ., Atlanta, GA

Abstract: Proteomic studies using postmortem human brain tissue samples have yielded robust assessments of the aging and neurodegenerative disease(s) proteomes. While these analyses

provide lists of molecular alterations in human conditions, like Alzheimer's disease (AD), identifying individual proteins that affect biological processes remains a challenge. To complicate matters, protein targets may be highly understudied and have limited information on their function. To address these hurdles, we sought to establish a blueprint to aid selection and functional validation of targets from proteomic datasets. A cross-platform pipeline was engineered to focus on synaptic processes in the entorhinal cortex (EC) of human patients, including controls, preclinical AD, and AD cases. Label-free quantification mass spectrometry (MS) data (n = 2260 proteins) was generated on synaptosome fractionated tissue from Brodmann area 28 (BA28; n = 58 samples). In parallel, dendritic spine density and morphology was measured in the same individuals. Weighted gene co-expression network analysis was used to construct a network of protein co-expression modules that were correlated with dendritic spine metrics. Module-trait correlations were used to guide unbiased selection of Twinfilin-2 (TWF2), which was the top hub protein of a module that positively correlated with thin spine length. Using CRISPR-dCas9 activation strategies, we demonstrated that boosting endogenous TWF2 protein levels in primary hippocampal neurons increased thin spine length, thus providing experimental validation for the human network analysis. Collectively, this study describes alterations in dendritic spine density and morphology as well as synaptic proteins and phosphorylated tau from the entorhinal cortex of preclinical and advanced stage AD patients.

Disclosures: K.M. Greathouse: None. **C.K. Walker:** None. **A.J. Weber:** None. **N.T. Seyfried:** None. **J.H. Herskowitz:** None.

Poster

PSTR129. Alzheimer's Disease: Molecular and Cellular Mechanisms I

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR129.06/D58

Topic:

Support: NINDS NS061788
NIA AG067635
NIA AG061800
NIA AG054719
NIA AG063755
NIA AG068024

Title: Dendritic spine head diameter predicts episodic memory performance in older adults

Authors: *J. H. HERSKOWITZ[1], C. K. WALKER[2], E. LIU[2], K. M. GREATHOUSE[2], D. A. BENNETT[3], N. T. SEYFRIED[4], C. GAITERI[5];
[1]Neurol., Univ. of Alabama at Birmingham, Birmingham, AL; [2]Neurol., Univ. of Alabama Birmingham, Birmingham, AL; [3]Rush Univ. Med. Ctr., Rush Univ. Med. Ctr., Chicago, IL; [4]Emory Sch. Med., Emory Univ., Atlanta, GA; [5]Psychiatry, SUNY, Syracuse, NY

Abstract: Episodic memory in older adults is varied and perceived to rely on numbers of synapses or dendritic spines. We analyzed spine density and morphology in temporal and premotor cortex from 128 older individuals. LASSO regression and nested model cross-validation revealed that dendritic spine head diameter in the temporal cortex, but not premotor cortex, improved prediction of episodic memory performance in models containing β-amyloid plaque scores, neurofibrillary tangle pathology, and sex. These findings support the emerging hypothesis that in the temporal cortex synapse strength is more critical than quantity for memory in old age.

Disclosures: J.H. Herskowitz: None. **C.K. Walker:** None. **E. Liu:** None. **K.M. Greathouse:** None. **D.A. Bennett:** None. **N.T. Seyfried:** None. **C. Gaiteri:** None.

Poster

PSTR129. Alzheimer's Disease: Molecular and Cellular Mechanisms I

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR129.07/D59

Topic:

Support: NIA T32NS095775
NIA R01 AG061800
NIA R01 AG054719
NIA R01 AG063755

Title: Tau-mediated synaptic dysfunction in Alzheimer’s disease

Authors: *A. J. WEBER, K. GREATHOUSE, E. LIU, J. HERSKOWITZ;
Univ. of Alabama at Birmingham, Birmingham, AL

Abstract: Amyloid-β (Aβ) plaques and neurofibrillary tangles (NFTs) of the microtubule-associated protein tau are the pathological hallmarks of Alzheimer’s disease (AD). In AD patients, the extent of NFT spread throughout the brain correlates with the severity of cognitive impairment. NFT burden is inversely correlated with dendritic spine loss, suggesting that tau contributes to synapse loss in AD. Additionally, transgenic mouse models of tauopathy demonstrate that tau is associated with dysfunctional neuronal activation. In this study, we investigated dendritic spine density and morphology as well as synaptic function in an *in vitro* model of tau accumulation. Human AD brain-derived tau paired helical filaments (AD-tau) were used to seed aggregation of endogenous rat tau in primary neuronal cultures. AD-tau was purified using differential centrifugation of sarkosyl-insoluble fractions. To begin understanding synaptic alterations, primary cortical rat neurons were seeded with AD-tau on multielectrode arrays (MEA) to measure local field potentials generated by spontaneous firing of neurons. MEA recordings were evaluated over a 23-day time course which assessed longitudinal changes in mean neuronal firing rate. To evaluate dendritic spine alterations, individual dendrites were imaged using wide-field microscopy. Neurolucida 360 was employed for three-dimensional

dendritic reconstructions and spine morphometric analysis. Our findings provide evidence that AD-tau induces endogenous tau accumulation, causing robust synaptic dysfunction and alterations in spine density and morphology.

Disclosures: **A.J. Weber:** None. **K. Greathouse:** None. **E. Liu:** None. **J. Herskowitz:** None.

Poster

PSTR129. Alzheimer's Disease: Molecular and Cellular Mechanisms I

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR129.08/D60

Topic: C.02. Alzheimer's Disease and Other Dementias

Title: Effects of a maternal high-fat diet during lactation in male and female offspring in a mouse model of tauopathy

Authors: ***T. GAUVRIT**[1], H. BENDERRADJI[1], K. CARVALHO[1], F. DELAHAYE[2], E. FAIVRE[1], A. LAUNAY[1], B. THIROUX[1], A. DELEAU[1], S. EDDARKAOUI[1], A. BOGDANOVA[1], M. BESEGHER[3], S. ABOULOUARD[4], S. LE GRAS[5], A. TAILLEUX[6], M. SALZET[4], L. BUÉE[1], D. BLUM[1], D. VIEAU[1];
[1]UMR-S1172 - LilNCog, Univ. de Lille, Inserm, CHU Lille, Lille, France; [2]Precision Oncology, Sanofi Oncology Res., Vitry-sur-Seine, France; [3]US 41-UMS 2014-PLBS, Animal Facility, Univ. de Lille, CNRS, Inserm, CHU Lille, Inst. Pasteur de Lille, Lille, France; [4]U1192 – Lab. PRISM, Univ. de Lille, Inserm, Lille, France; [5]CNRS U7104, Inserm U1258, GenomEast Platform, IGBMC, Univ. de Strasbourg, Illkirch, France; [6]U1011 EGID, Univ. de Lille, Inserm, CHU Lille, IPL, Lille, France

Abstract: The perinatal environment has been suggested to participate to the development of tauopathies and Alzheimer’s disease but the molecular and cellular mechanisms involved remain contradictory and under-investigated. Here, we evaluated the effects of a maternal high-fat diet (mHFD) during lactation on the development of tauopathy in the THY-Tau22 mouse strain, a model of progressive Tau pathology associated with cognitive decline. During lactation, dams were fed either a standard diet (13.6% fat) or a mHFD (58% fat). At weaning (postnatal day 21), offspring was fed a standard diet until sacrifice at 4 months of age (i.e. the onset of Tau pathology) or 7 months of age (i.e. the onset of cognitive impairment). During lactation, the mHFD promotes an increased body weight gain in offspring. At 3 months of age, the mHFD leads to a glucose intolerance only in male offspring. The mHFD increased hippocampal Tau pathology at 4 months of age in males and at 7 months of age in females. Altogether, these data suggest a sexual dimorphism with the male offspring being impacted earlier. These changes led to impaired spatial memory at 7 months of age in both sexes. In the hippocampus, using omics approaches (RNA sequencing and mass spectrometry), we showed that the mHFD increases the number of genes and proteins deregulated with the development of Tau pathology and modifies gene expression and protein levels in a sex-dependent manner. Interestingly, although these deregulated genes and proteins are different, there are enriched in similar cell components such

as synaptic compartments and mitochondria in both sexes, and in the neurogenesis process only in males. Moreover, among the deregulated proteins, calretinin, a marker of immature neurons, was found to be decreased by mHFD in males with age. Taken together, our data showed for the first time that mHFD has long-lasting and sex-dependent effects on the development of tauopathy and reinforced the idea that age-related neurodegenerative diseases may have, at least in part, a neurodevelopmental origin.

Disclosures: **T. Gauvrit:** None. **H. Benderradji:** None. **K. Carvalho:** None. **F. Delahaye:** None. **E. Faivre:** None. **A. Launay:** None. **B. Thiroux:** None. **A. Deleau:** None. **S. Eddarkaoui:** None. **A. Bogdanova:** None. **M. Besegher:** None. **S. Aboulouard:** None. **S. Le Gras:** None. **A. Tailleux:** None. **M. Salzet:** None. **L. Buée:** None. **D. Blum:** None. **D. Vieau:** None.

Poster

PSTR129. Alzheimer's Disease: Molecular and Cellular Mechanisms I

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR129.09/D61

Topic: C.02. Alzheimer"s Disease and Other Dementias

Support: ADORASTRAU, ANR Grant

Title: Effects of adenosine A2A receptor astrocytic upregulation in the mouse hippocampus

Authors: *A. LAUNAY[1], K. CARVALHO[1], T. GAUVRIT[1], B. THIROUX[1], D. VIEAU[1], E. AUGUSTIN[2], K. CAMBON[2], A. BEMELMANS[2], G. BONVENTO[2], S. LE GRAS[3], J.-S. ANNICOTTE[4], L. BUÉE[1], E. FAIVRE[1], D. BLUM[1];
[1]UMR-S1172 - LilNCog, Univ. de Lille, Inserm, CHU Lille, Lille, France; [2]CEA,CNRE, MIRCen, Neurodegenerative Dis. Lab., Fontenay-aux-roses, France; [3]GenomEast Platform, Inst. de Génétique et de Biologie Moléculaire (IGBMC), Strasbourg, France; [4]Univ. Lille, Inserm, CHU Lille, Inst. Pasteur de Lille, U1167—RID-AGE—Facteurs de risque et déterminants moléculaires des maladies liées au vieillissement, Lille, France

Abstract: Alzheimer's disease (AD) is notably characterized by the intraneuronal aggregation of tau proteins which play an important role in synaptic dysfunctions and memory decline. Studies have reported that chronic caffeine consumption reduces AD risk and cognitive deficits. These protective effects would be ascribed to the blockade of adenosine A_{2A} receptors (A_{2A}Rs) which are pathologically upregulated in the hippocampus of patients with AD. This upregulation is observed in the astrocytes -which are essential cells for brain homeostasis- and has been correlated with the pathology development and associated cognitive deficits. However, the mechanisms underlying the link between astrocytic A_{2A}R upregulation and memory deficits remain unclear. To uncover the impact of astrocytic A_{2A}R upregulation, we intrahippocampally injected an AAV2/9 virus expressing A_{2A}R (AAV-A2A), or GFP as control (AAV-GFP), under a GFAabc1d astrocyte-specific promoter, in 2m-old C57Bl6/J mice. We evaluated consequences in

term of spatial memory performance, response to hippocampal neural networks using DREADDs as well as astrocyte reactivity, morphology and transcriptome. Our data show that $A_{2A}R$ overexpression in hippocampal astrocytes impairs short-term spatial memory (Y-Maze task) and long-term spatial learning (Barnes Maze task). At the network level, thanks to the DREADD approach, we observed an enhanced neuronal excitability in animals injected with the AAV-A2A as compared to the control GFP group, characterized by a higher immediate early gene response. These changes were associated with deep alterations of astrocyte reactivity, morphology and transcriptome. These results therefore demonstrate that upregulation of $A_{2A}R$ in hippocampal astrocytes, as seen in the brains of AD patients, is sufficient to alter astrocytic phenotype, neuronal response and memory. To determine the pathophysiological impact of $A_{2A}R$ astrocytic upregulation, we are now currently determining the effects of the latter at an early stage using a mouse model of AD-like tauopathy (Thy-Tau22). Our first results indicate that $A_{2A}R$ dysregulation in astrocytes potentiates Tau-induced memory deficits. Impact on brain lesions as well as on glial response are currently under investigation. We expect to uncover mechanisms linking astrocyte dysfunction to the evolution of tauopathy and provide additional proof-of-concept that targeting $A_{2A}R$ is of therapeutical importance in AD and tauopathies.

Disclosures: **A. Launay:** None. **K. Carvalho:** None. **T. Gauvrit:** None. **B. Thiroux:** None. **D. Vieau:** None. **E. Augustin:** None. **K. Cambon:** None. **A. Bemelmans:** None. **G. Bonvento:** None. **S. Le Gras:** None. **J. Annicotte:** None. **L. Buée:** None. **E. Faivre:** None. **D. Blum:** None.

Poster

PSTR129. Alzheimer's Disease: Molecular and Cellular Mechanisms I

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR129.10/D62

Topic: C.02. Alzheimer"s Disease and Other Dementias

Support: Fondation pour la Recherche Médicale
Fondation Alzheimer
ANR
Région Hauts de France
Labex DISTALZ
Inserm
Université de Lille

Title: Neuronal adenosine A_{2A} receptor upregulation exacerbates memory deficits and associated synaptic loss in an amyloidogenic mouse model of Alzheimer’s disease.

Authors: *E. FAIVRE[1], V. GOMEZ-MURCIA[1], A. LAUNAY[1], K. CARVALHO[1], C. MONTMASSON[2], C. MERIAUX[1], B. THIROUX[1], R. CAILLIEREZ[1], M. BESEGHER[3], S. BEGARD[1], M. WISZTORSKI[4], I. FOURNIER[4], N. DEGLON[5], A.-P. BEMELMANS[6], M. HAMDANE[1], S. LEVI[2], D. VIEAU[1], L. BUÉE[1], D. BLUM[1];

[1]UMR-S1172 - LilNCog, Univ. de Lille, Inserm, CHU Lille, Lille, France; [2]Inserm, U1270, Inst. du Fer à Moulin, team Plasticity in cortical network and epilepsy, Paris, France; [3]UAR2014 - US41- Plateformes Lilloises en Biologie & Santé, Lille, France; [4]Inserm U1192, Univ. de Lille, Lab. PRISM, Lille, France; [5]Neurosciences Res. Center, Lab. of Cell. and Mol. Neurotherapies, Lausanne, Switzerland; [6]Univ. Paris-Saclay, CEA, CNRS, Lab. des Maladies Neurodégénératives : mécanismes, thérapies, imagerie, Fontenay-aux-Roses, France

Abstract: Epidemiological and experimental studies pointed-out that chronic caffeine consumption reduces Alzheimer's Disease (AD) risk and associated cognitive deficits. These protective effects are thought to be ascribed to the blockade of adenosine A_{2A} receptors (A_{2A}Rs). Interestingly, the latter are found abnormally upregulated in neurons of AD patient's brains in correlation with the development of cognitive deficits. These observations suggest a link between neuronal A_{2A}R dysregulation and memory impairments in AD. To get insights into the impact of A_{2A} dysregulation in AD, we evaluated the consequences of a neuronal A_{2A}R upsurge in a transgenic model of AD-like amyloidogenesis (APP/PS1dE9 mice). We crossed APP/PS1 mice with the transgenic TRE-A_{2A} strain, carrying the mouse A_{2A}R under the control of a Tet-responsive-element promoter. This led rise to four genotypic groups: WT, APP, WT/TRE-A_{2A} and APP/TRE-A_{2A}. At 3m of age, all the animals were bilaterally injected in the hippocampus with an AVV2/5-CBA-ttA, allowing the expression of ttA transactivator and then neuronal A_{2A}R upsurge in TRE-A_{2A} animals. At 6m of age, when APP/PS1 mice do not display deficits, behavioral evaluations revealed that neuronal A_{2A}R overexpression strongly worsens spatial memory impairments of APP animals. While that was not associated with significant change in amyloid burden, neuronal A_{2A}R upsurge favored the development of neuritic tau pathology. Mass spectrometry-based high-throughput proteomics identified modifications in the molecular profile of APP/TRE-A_{2A}. Gene ontology analysis revealed that proteins the most highly differentially downregulated were related to synaptic function. Immunohistochemistry and western blot analysis confirmed loss of pre and postsynaptic markers when neuronal A_{2A}R is upregulated in APP mice. These data support that pathological upregulation of A_{2A}R in neurons enhance synapse vulnerability to amyloid and is instrumental towards the decline of cognitive functions in AD.

Disclosures: E. Faivre: None. **V. Gomez-Murcia:** None. **A. Launay:** None. **K. carvalho:** None. **C. Montmasson:** None. **C. Meriaux:** None. **B. Thiroux:** None. **R. Caillierez:** None. **M. Besegher:** None. **S. Begard:** None. **M. Wisztorski:** None. **I. Fournier:** None. **N. Deglon:** None. **A. Bemelmans:** None. **M. Hamdane:** None. **S. levi:** None. **D. vieau:** None. **L. Buée:** None. **D. Blum:** None.

Poster

PSTR129. Alzheimer's Disease: Molecular and Cellular Mechanisms I

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR129.11/D63

Topic: C.02. Alzheimer"s Disease and Other Dementias

Support: LabEx DISTALZ
ANR ADORAsTRAU
ANR EPIFUS
France Alzheimer
AFI
FCT

Title: Caffeine intake exerts dual genome-wide effects on hippocampal metabolism and learning-dependent transcription

Authors: *D. BLUM[1], I. PAIVA[2], O. NEBIE[1], C. MÜLLER[3], E. FAIVRE[1], D. VIEAU[1], L. V. LOPES[4], L. BUEE[1], R. A. CUNHA[5], J.-S. ANNICOTTE[1], A.-L. BOUTILLIER[2];
[1]Inserm, Lille, France; [2]LNCA, Strasbourg, France; [3]Univ. of Bonn, Bonn, Germany; [4]Inst. de Medicina Molecular, Fac Med. Lisbon, Inst. de Medicina Mol. João Lobo Antunes, Fac Med. Lisbon, Lisbon, Portugal; [5]Univ. of Coimmbra, Univ. of Coimbra, Coimbra, Portugal

Abstract: Caffeine is the most widely consumed psychoactive substance in the world. Strikingly, the molecular pathways engaged by its regular consumption remain unclear. We herein addressed the mechanisms associated with habitual (chronic) caffeine consumption in the mouse hippocampus using untargeted orthogonal omics techniques. Our results revealed that chronic caffeine exerts concerted pleiotropic effects in the hippocampus at the epigenomic, proteomic, and metabolomic levels. Caffeine lowered metabolism-related processes (e.g., at the level of metabolomics and gene expression) in bulk tissue, while it induced neuron-specific epigenetic changes at synaptic transmission/plasticity-related genes and increased experience-driven transcriptional activity. Altogether, these findings suggest that regular caffeine intake improves the signal-to-noise ratio during information encoding, in part through fine-tuning of metabolic genes, while boosting the salience of information processing during learning in neuronal circuits.

Disclosures: D. Blum: None. **I. Paiva:** None. **O. Nebie:** None. **C. Müller:** None. **E. Faivre:** None. **D. Vieau:** None. **L.V. Lopes:** None. **L. Buee:** None. **R.A. Cunha:** None. **J. Annicotte:** None. **A. Boutillier:** None.

Poster

PSTR129. Alzheimer's Disease: Molecular and Cellular Mechanisms I

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR129.12/D64

Topic: C.02. Alzheimer's Disease and Other Dementias

Support: P20-GM109036

Title: Characterization of overexpressing neuronal-specific miR-34a animal model for neurodegeneration

Authors: *I. PURSELL[1], R. FREITAS[2], L. GARFINKEL[2], M. MARCUS[2], H. WANG[2], E. ENGLER-CHIURAZZI[2];
[2]Neurosurg., [1]Tulane Univ., New Orleans, LA

Abstract: Alzheimer's Disease (AD) is a common age-related neurodegenerative condition causing dementia. Despite extensive research, the underlying factors and genetic role in AD progression remain unclear. MicroRNAs, small RNA molecules regulating protein expression, contribute to neurodegeneration. Specifically, microRNA-34a (miR-34a) is a key regulator of neurological function associated with neurodegenerative disorders. However, there is a lack of effective tools to study miR-related neurodegeneration *in vivo*.In our previous research, mice overexpressing miR-34a ubiquitously and treated with doxycycline (Doxy) displayed AD-related neuropathologies. We aimed to generate a neuronal-specific mouse model with inducible miR-34a overexpression to investigate the consequences of elevated miR-34a levels in aging. We hypothesized that miR-34a overexpression would lead to a dementia-like phenotype characterized by cognitive impairments and the accumulation of AD-related neuropathological features. We investigated the cognitive and neurobiological consequences of miR-34a overexpression *in vivo* using a Doxy-inducible miR-34a expression mouse model, restricting miR-34a overexpression to excitatory neurons through the CaMKIIα driver. M/F miR-34a+/- mice aged 28-46 weeks were treated with water or Doxy (2mg/ml) for 7 or 75-90 days to induce miR-34a expression. We evaluated cognitive and neurobiological factors known to be disrupted in other transgenic AD mouse models.We found a decrease in the expression of miR34a target genes (NR2B & Shank2) and elevated levels of miR34a-5p were found in the hippocampus of overexpressing mice compared to peripheral tissues like the kidney. In addition, mice displayed decreased percent spontaneous alternations compared to control animals. We initiated the optimization of isolating distinct nervous system cell populations for miRNA analysis to investigate region-specific changes in miR34a levels.In summary, our inducible global miR-34a overexpression mouse model exhibited several characteristics of a dementia phenotype. Although our excitatory neuron-specific miR-34a overexpression model requires further refinement, initial findings revealed modest increases in hippocampal miR34a levels associated with slight reductions in synaptic targets and subtle cognitive changes. This suggests that the AD phenotype induced in our global miR34a overexpression model may involve contributions from other neural or peripheral cell types. The reversible and targetable nature of our model offers valuable insights into the role of miR-34a in the progression of AD.

Disclosures: I. Pursell: None. **R. Freitas:** None. **L. Garfinkel:** None. **M. Marcus:** None. **H. Wang:** None. **E. Engler-Chiurazzi:** None.

Poster

PSTR129. Alzheimer's Disease: Molecular and Cellular Mechanisms I

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR129.13/D65

Topic: C.02. Alzheimer's Disease and Other Dementias

Support: NIH Grant R01NS089578
DOD W81XQH-18-1-0338

Title: Loss of Par1b/MARK2 results in increased neuronal excitability and dysregulation of transcriptional programs associated with ion homeostasis and synaptic transmission

Authors: *H. CAIOLA[1], Q. WU[4], S. SONI[4], K. MONAHAN[2], D. MARGOLIS[3], D. CROCKETT[4], H. ZHANG[4];
[2]Mol. Biol. and Biochem., [3]Cell Biol. and Neurosci., [1]Rutgers Univ., Piscataway, NJ; [4]Neurosci. and Cell Biol., Rutgers RWJMS, Piscataway, NJ

Abstract: Neurodegenerative disorders are a major cause of death and disability in the United States and are increasing in prevalence at an alarming rate. Par1/MARK serine threonine kinases have been genetically linked to neurodegenerative diseases such as Alzheimer's disease (AD). In addition, we and others have found that human AD patients have decreased levels of Par1b/MARK2 in the medial temporal lobe. Furthermore, we found that loss of Par1b/MARK2 in mice results in AD-related phenotypes such as memory impairments, age-dependent cortical and hippocampal degeneration, and synaptic dysfunction. Par1b/MARK2 is known to function upstream of several transcriptional regulators implicated in AD. Moreover, our RNAseq analysis in Par1b/MARK2 knockouts suggests that programs associated with synaptic transmission and ion homeostasis are highly dysregulated. This is interesting given that dysregulation of ion channels is highly associated with seizures, which has been proposed as an early biomarker of AD. Indeed we found that heterozygous knockout of Par1b/MARK2 in mice results in increased seizure susceptibility, suggesting underlying neuronal hyperexcitability. Current efforts aim to use *in vivo* calcium imaging to determine whether loss of Par1b/MARK2 results in increased spontaneous calcium transients in knockout mice. Together, these data will help us understand how Par1b/MARK2 contributes to the regulation of neuronal excitability, which could give insight into potential mechanisms for Par1b/MARK2 in neurodegeneration and AD.

Disclosures: H. Caiola: None. **Q. Wu:** None. **S. Soni:** None. **K. Monahan:** None. **D. Margolis:** None. **D. Crockett:** None. **H. Zhang:** None.

Poster

PSTR129. Alzheimer's Disease: Molecular and Cellular Mechanisms I

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR129.14/D66

Topic: C.02. Alzheimer"s Disease and Other Dementias

Support: NIH R01AG057555

Title: Potential of targeting alpha-1 adrenergic receptor and phosphoglycerate kinase 1 with Terazosin to treat Alzheimer's disease using a transgenic AD rat model

Authors: *S. PATTANASHETTY GANGADHARAPPA[1,3], P. ROCKWELL[1,3], P. SERRANO[1,5], L. XIE[2,4], M. FIGUEIREDO-PEREIRA[1,3];
[1]Dept. of Biol., [2]Dept. of Computer Sci., Hunter College, CUNY, New York, NY; [3]Biol. PhD Program, [4]Biochem. PhD program, The Grad. Ctr., New York, NY; [5]Dept. of Psychology, Hunter college, CUNY, New York, NY

Abstract: Alzheimer's disease (AD) is a neurodegenerative disorder without a cure. Targeting this multifactorial disease by repurposing FDA approved drugs serves as a faster mode of treatment due to its pre-established human safety. We tested terazosin (TZ), an α-1 adrenergic receptor (AR) antagonist and phosphoglycerate kinase-1 (PGK1) activator as having potential to treat AD. Early stage of AD is characterized with loss of noradrenergic neurons in the locus coeruleus (LC) that initiates an over compensatory mechanism leading to increased production of norepinephrine. This leads to increased synaptic activity in LC thus increasing extracellular Aβ deposition via endocytosis of APP. Furthermore, dysregulated glycolysis causing decreased ATP levels is shown to play a causal role in the progression of AD and PGK1 is the first ATP generating enzyme in glycolysis. We hypothesize that TZ via its dual mechanism of action by blocking α-1 AR and activating PGK1, will exert neuroprotective effects and ameliorate AD pathology by decreasing the extracellular Aβ deposition and increasing ATP levels respectively. To test this, we used the TgF-344AD rat model that develops AD in an age dependent manner. TZ treatment (0.5 mg/kg of body weight/day) starts at 5 months of age (pre pathology) until 11 months of age (moderate pathology). At 9 and 11 months of age, we used the active place avoidance test to compare the hippocampal-dependent spatial and working memory of the transgenic not treated (TGNT) and transgenic treated (TGTR) rats with terazosin. Wild type rats were also tested as a control. Our data indicate that TZ significantly improves spatial and working memory of TGTR rats while TGNT show loss of hippocampal-dependent memory. This indicates the potential of TZ to treat AD. Additionally, preliminary data on immunohistochemical analysis appears to show beneficial effects of TZ in decreasing both Aβ plaques and neuroinflammation in TGTR rats. However, further analysis is required to confirm statistical significance. Future studies will perform immunocytochemistry, RNAseq, western blots, and luciferase assay on the treated and untreated Tg-AD groups to test the effects of TZ on other AD pathologies. Overall, our studies will decipher the potential of TZ to treat AD and present new molecular candidates that can be targeted to cure or delay the progression of AD.
Supported by NIH R01AG057555 [to L.X. (PI), M.E.F.-P., P.R., P.S. (co-Is)] and the City University of New York (MCD program, Graduate Center).

Disclosures: S. Pattanashetty Gangadharappa: None. **P. Rockwell:** None. **P. Serrano:** None. **L. Xie:** None. **M. Figueiredo-Pereira:** None.

Poster

PSTR129. Alzheimer's Disease: Molecular and Cellular Mechanisms I

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR129.15/D67

Topic: C.02. Alzheimer's Disease and Other Dementias

Support: Jeanne B. Kempner Scholarship (2022-2023, C.N.)
AARG-17-533363 (B.K.)
NIA R21 – AG059223 (B.K.)
NIA R01 – AG063945 (B.K.)
The Don and Nancy Mafrige Professor in Neurodegenerative Disease Endowment (B.K.)
Mitchell Center for Neurodegenerative Diseases

Title: Elucidating the role of Phospholipase D1 (PLD1) in neurodegenerative states using post-mortem human brain samples

Authors: *S. BUDHWANI, S. SREENIVASA MURTHY, C. NATARAJAN, K. GARZA, J. DORNAK, B. KRISHNAN;
Neurol., Univ. of Texas Med. Br., Galveston, TX

Abstract: Title: Elucidating the role of Phospholipase D1 (PLD1) in neurodegenerative states using post-mortem human brain samplesAuthors: Shaneilahi Budhwani, Sravan Gopalkrishna Shetty Sreenivasa Murthy, Chandramouli Natarajan, Klarissa Garza, Jared Dornak, Balaji KrishnanBackground: A lipolytic enzyme called phospholipase D (PLD) predominantly breaks down membrane phospholipids. There are two mammalian isoforms of the phosphatidylcholine (PC)-specific PLD - PC-PLD1 and PC-PLD2. As a result of its conserved status, this enzyme participates in numerous signaling pathways, including synaptic neurotransmission, which was demonstrated to be essential in the acquisition and expression of associative memory during normal healthy states. However, only PLD1 expression is aberrantly elevated in human post-mortem brain regions from Alzheimer's Disease (AD) or behavioral variant Frontotemporal Dementia (bvFTD) patients. **In the present study**, we are hypothesizing that elevated PLD1 levels impinge on mechanisms of autophagy and neuroinflammation to accelerate the process of cognitive decline in neurodegenerative states. We are elucidating this association by establishing human clinical relevance of PLD1 using the markers of astrocytic changes (GFAP) and neuroinflammation (IBA1, CD68) among others. We report an increased co-localization of PLD1 with these markers. Other biochemical assessments of the profile are in progress and will provide insight into the signaling events that are altered by PLD1. **Methods:** Immunofluorescence and other biochemical approaches including Western blotting, immunoprecipitation, RNAseq, and Proteomics were used to study human post-mortem brain samples. **Acknowledgements:** Jeanne B. Kempner Scholarship (2022-2023, C.N.), AARG-17-533363 (B.K.), NIA R21 – AG059223 (B.K.), NIA R01 – AG063945 (B.K.), The Don and Nancy Mafrige Professor in Neurodegenerative Disease Endowment (B.K.), Mitchell Center for Neurodegenerative Diseases

Disclosures: S. Budhwani: None. **S. Sreenivasa Murthy:** None. **C. Natarajan:** None. **K. Garza:** None. **J. Dornak:** None. **B. Krishnan:** None.

Poster

PSTR129. Alzheimer's Disease: Molecular and Cellular Mechanisms I

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR129.16/D68

Topic: C.02. Alzheimer's Disease and Other Dementias

Support: NIH Grant AG077610
NIH Grant AG079141

Title: Characterization of cell and region specific human BIN1 overexpression in a transgenic mouse model

Authors: *N. RAM, S. WANG, M. PONNUSAMY, M. YUKSEL, M. DELOZIER, V. SKOROBOVENKO, S. SMIRNOU, L. COLLIER, G. THINAKARAN;
Mol. Med., Univ. of South Florida Morsani Col. of Med., Tampa, FL

Abstract: Genome-wide association studies have identified the bridging integrator 1 gene (*BIN1)* as the second leading genetic risk factor for late-onset Alzheimer's Disease (LOAD). The gene encodes for over 20 isoforms of the adaptor protein that are expressed in a tissue- and cell-type-specific manner. These isoforms serve diverse roles in various cell types, including membrane remodeling, endocytosis, and neurotransmitter vesicle release. BIN1 has also been described to participate in tau and amyloid pathologies that are hallmarks of AD. However, the specific role of BIN1 in AD pathogenesis is not fully understood. Patients with LOAD have altered expression of alternatively spliced BIN1 isoforms, notably decreased neuronal isoforms and increased ubiquitous isoforms. Our goal is to study the cell-type-specific expression of human *BIN1* isoforms in a transgenic mouse model to understand better its function and, eventually, its role in AD pathogenesis and neurodegeneration. A transgenic mouse line, *B6 Tg(Bin1)U154.16.16Yah* (referred to as Tg^{Hb}), which has 5-10 copies of the entire *BIN1* locus and upstream sequences contained in a ~195 kb human BAC, was recently reported. The cell-type-specific and region-specific BIN1 expression have not yet been studied in this model. Here, we examined BIN1 expression in the Tg^{Hb} line via immunoblotting, immunohistochemistry, and other biochemical techniques. Our results indicate a significant increase in BIN1 protein expression in both the gray matter and white matter of transgenic mouse brains compared to non-transgenic controls. The Tg^{Hb} transgenic line has a significant increase in the abundance of neuronal and non-neuronal BIN1 isoforms in the brain. Our ongoing investigation aims to ascertain whether the overexpression of human BIN1 will modulate AD pathogenesis and neurodegeneration.

Disclosures: N. Ram: None. **S. Wang:** None. **M. Ponnusamy:** None. **M. Yuksel:** None. **M. Delozier:** None. **V. Skorobovenko:** None. **S. Smirnou:** None. **L. Collier:** None. **G. Thinakaran:** None.

Poster

PSTR129. Alzheimer's Disease: Molecular and Cellular Mechanisms I

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR129.17/D69

Topic: C.02. Alzheimer's Disease and Other Dementias

Support: NIH RF1 AG059405
NIH T32 NS061788
NIH T32 HD071866

Title: Loss of Alzheimer's disease risk factor BIN1 from inhibitory neurons induces network hyperexcitability

Authors: *N. DAVIS[1], Y. VOSKOBIYNYK[2], N. COCHRAN[2], E. D. ROBERSON[2];
[2]Neurol., [1]Univ. of Alabama, Birmingham, Birmingham, AL

Abstract: Alzheimer's disease (AD) is the most common neurodegenerative disease, affecting more than 6 million Americans. Despite its prevalence, much is still not understood about the disease. To better understand the disease, genome-wide association studies have been conducted to identify genetic risk factors. One risk factor, a single nucleotide polymorphism in the *bridging integrator 1 (BIN1)* gene, is present in approximately 40% of the population and has the largest effect size of the common AD genetic risk factors. While the association between *BIN1* and AD has been established, the function of the protein and its contribution to AD remains understudied. We previously showed that increasing *BIN1* expression in primary hippocampal neurons increases neuronal excitability (Voskobiynyk & Roth et al., 2020). However, primary hippocampal neurons are predominantly composed of excitatory neurons, so the contribution of BIN1 from inhibitory neurons remains unknown. This is particularly important as inhibitory dysfunction contributes to AD, affecting cognitive function and epileptiform activity. Additionally, expression of the primary neuronal isoform of BIN1 is lower in AD patients compared to healthy age-matched controls. We thus hypothesized that a loss of Bin1 from all neurons will lead to increased network hyperexcitability, with differing effects with a loss of Bin1 from excitatory or inhibitory neurons. Using conditional knockout mice, we selectively reduced murine Bin1 from all neurons (*Nestin*-Cre-driven), excitatory neurons (*CaMKIIα*-Cre-driven), or inhibitory neurons (*Viaat*-Cre-driven). We examined network hyperexcitability through a pentetrazol-induced seizure susceptibility assay and found that a pan-neuronal loss of Bin1 increased seizure susceptibility in a gene-dose dependent manner. Bin1 loss from excitatory neurons decreased seizure susceptibility, while Bin1 loss from inhibitory neurons increased seizure susceptibility, suggesting that it is the loss of Bin1 from inhibitory neurons driving the overall effect. We then generated a mouse line to selectively reduce Bin1 in parvalbumin (PV) interneurons to further define cell-type specificity. While PV interneurons contribute to inhibitory dysfunction in AD, we saw no change in seizure susceptibility from controls. This suggests that a different interneuron subpopulation may be responsible for the changes in seen in the total inhibitory knockout of Bin1, or that loss of Bin1 from PV interneurons alone is not sufficient to drive this overall effect. Overall, this study contributes to our understanding of how BIN1 regulates network hyperexcitability at a cell-type specific level.

Disclosures: N. Davis: None. **Y. Voskobiynyk:** None. **N. Cochran:** None. **E.D. Roberson:** F. Consulting Fees (e.g., advisory boards); AGTC, Lilly, Genentech. Other; Editorial board of Journal of Neuroscience.

Poster

PSTR129. Alzheimer's Disease: Molecular and Cellular Mechanisms I

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR129.18/Web Only

Topic: C.02. Alzheimer's Disease and Other Dementias

Support: PAPIIT-DGAPA IN216821

Title: Vanadium pentoxide (v_2o_5) inhalation as an induced model of Alzheimer's disease

Authors: *C. DORADO-MARTÍNEZ[1], E. MONTIEL-FLORES[2], J. ORDONEZ LIBRADO[2], A. GUTIÉRREZ-VALDEZ[2], C. GARCIA-CABALLERO[2], J. ESPINOSA-VILLANUEVA[2], L. REYNOSO-ERAZO[2], M. AVILA-COSTA[2];
[1]UNAM, Ciudad DE Mexico, Mexico; [2]UNAM, TLALNEPANTLA, Mexico

Abstract: Alzheimer's disease (AD) is the most common neurodegenerative pathology worldwide, it has been reported that approximately 15 million people suffer from this disease, the incidence annually increases 0.5% in 65-year-old people and 8% in 85-year-olds; it has been difficult to find an animal model that replicates all the characteristics of the AD neurodegenerative process. Previous experiments in our laboratory have shown that chronic exposure to vanadium pentoxide (V2O5) in rats causes morphological and behavioral changes similar to those seen in AD. To this end 40 male Wistar rats were randomly divided into two control and two experimental groups (n = 10). The experimental groups were exposed to V2O5 0.02M for 1 h, 3 times a week, for 6 months, after 6-month exposure one experimental group was leaved in a 6-month recovery phase. To measure behavioral changes, the four groups were trained in the T-maze test that assesses spatial MEMORY and an open field test for 10 mins. All groups were evaluated once a month for 6 or twelve months. To measure histological alterations, after 6 or 12 months, frontal and entorrinal cortex, CA1, subiculum and amygdala, underwent Congo red or argentic Bielschovsky impregnation and were analyzed. Memory results in the T-maze show memory impairment since the group had been exposed for three months to V2O5. Chronic V2O5 inhalation is a model of induced AD that causes alterations in spatial memory and motor behavior, accumulation of βA in the vascular endothelium and pyramidal neurons of the frontal and entorhinal cortex and accumulation of intraneuronal NFTs in pyramidal neurons of CA1 and subiculum and loss of dendritic spines, which allows us to propose a model that relates clinical manifestations with histological changes. Being an induced model represents sporadic AD which, although it represents more than 95% of cases. This model represents neurodegenerative changes specific to AD in which we observe production and accumulation of amyloid and plaques first in CA1, subiculum, enthorrinal cortex, amygdala and frontal cortex, production, and accumulation of NFTs, and spatial memory damage. There is a decrease in the number of neurons in the cortex and CA1, these alterations are not reversible after six months, indicating that once the neurodegenerative process is established and homeostasis is broken, the damage increases. Our model is compatible with Braak stage IV of AD, which represents a moment where it is feasible to propose therapies that have a positive impact on stopping neuronal damage.

Disclosures: **C. Dorado-Martínez:** None. **E. Montiel-Flores:** None. **J. Ordonez Librado:** None. **A. Gutiérrez-valdez:** None. **C. Garcia-Caballero:** None. **J. Espinosa-villanueva:** None. **L. Reynoso-Erazo:** None. **M. Avila-Costa:** None.

Poster

PSTR130. Vascular Changes in Alzheimer's Disease

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR130.01/D70

Topic: C.02. Alzheimer's Disease and Other Dementias

Support: National Cancer Institute R00CA214523
National Institute of Biomedical Imaging and Bioengineering R21EB030072
National Institute on Deafness and Other Communication Disorders R21DC019473
National Institute on Aging of the National Institutes of Health R03AG059103
Kiwanis Neuroscience Research Foundation
Beckman Institute Postdoctoral Fellowships

Title: Super resolution ultrasound imaging of progressive vascular abnormalities in 5xFAD mouse model of Alzheimer's Disease

Authors: *N. VAITHIYALINGAM CHANDRA SEKARAN[1,2], M. R. LOWERISON[2,3], P. SONG[2,3], D. A LLANO[2,4,5];
[1]Univ. of Illinois Urbana-Champaign, Urbana, IL; [2]Beckman Inst., [3]Dept. of Electrical & Computer Engin., [4]Mol. and Integrative Physiol., [5]Carle Illinois Col. of Med., Univ. of Illinois at Urbana-Champaign, Urbana, IL

Abstract: It is known that Vascular dysfunction and structural vascular abnormalities in Alzheimer's Disease (AD) tend to contribute to pathology progression, but less is known about the early role of the vasculature in AD progression. Studies of the impact of cerebrovascular disease on AD are complicated by the difficulty of imaging deep-brain structures with high fidelity. In the present study, we utilized super-resolution ultrasound localization microscopy which reveals microvascular functional and structural features throughout the whole brain depth. We used 3-and 6-month-old 5X FAD mouse models of AD with age-matched wild-type controls to study microvascular dynamics. The ULM cross-sectional images are co-registered with histological vascular staining and amyloid-beta deposition staining. We examined global hippocampal, entorhinal cortex, and isocortex alterations in microvascular density and median blood flow velocity from these two cohorts of animals. We found that functional decreases in hippocampal and entorhinal flow velocity preceded structural deficits in regional vascular density. Analysis of local blood profiles in the cortex and in lateral arteries further demonstrated a consistent flow velocity reduction in the 5xFAD group for specific blood vessels. We also

found that flow velocity decreases with increasing vascular tree order. In this study using the ULM confirming with histology we evidenced that microvascular functional changes precede microvascular structural changes and are most prominent in the hippocampus and entorhinal cortex.
Keywords: Microvasculature, Ultrasound localization microscopy, Alzheimer's disease, 5x FAD, Hippocampus.

Disclosures: N. Vaithiyalingam Chandra Sekaran: None. **M. R. Lowerison:** None. **P. Song:** None. **D. A Llano:** None.

Poster

PSTR130. Vascular Changes in Alzheimer's Disease

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR130.02/E1

Topic: C.02. Alzheimer's Disease and Other Dementias

Title: Mri and functional ultrasound reveal early pathogenic events in a dahl ss rat model of cerebrovascular disease

Authors: *A. BIALAS[1], R. IMMONEN[3], J. UHARI-VÄÄNÄNEN[3], H. VAHERTO[3], T. MIETTINEN[3], A. SHATILLO[3], J. DODGE[2];
[2]Rare and Neurologic Dis. Res. Therapeut. Area, [1]Sanofi, Cambridge, MA; [3]Charles River Discovery Services, Kuopio, Finland

Abstract: Regulation of cerebral blood flow is critical for healthy brain function. Cerebrovascular diseases, such as stroke, intracerebral hemorrhages, and vascular dementia, cause damage to the brain by altering cerebral blood flow. Furthermore, cerebrovascular dysfunction has emerged as a major risk factor and key player in the pathogenesis of neurodegenerative diseases. Thus, the mechanisms underlying cerebrovascular disease progression are an attractive therapeutic target; however, because ischemia and brain hemorrhages often occur without warning, understanding the earliest events in sporadic cerebrovascular disease has been challenging. The focus of this study was to use state-of-the art magnetic resonance imaging (MRI) and functional ultrasound (fUS) to characterize the onset and progression of cerebrovascular disease in the Dahl salt-sensitive (Dahl SS) rat model. The Dahl SS model develops hypertension, a common risk factor for cerebrovascular disease, with exposure to a high salt diet and is a well-established stroke model. To allow us to capture early events in cerebrovascular disease in this model, rats were fed a moderately high salt diet (4%) starting at 5 weeks of age and were monitored using after 2, 4, 6 and 8 weeks of salt loading. Experiments were performed on total of 50 Dahl-SS rats on salt diet while 12 Spraque Dawley (CD) rats on normal diet served as controls. Separate groups were imaged at each time point. MRI data were collected on 11.7T high field system (Bruker BioSpec). T2 weighted images and quantitative T2 maps were analyzed for edema or lesions, T2star images were screened for microbleeds, and Gadolinium contrast enhanced imaging for BBB disruptions. Terminal fUS

imaging was done after skull thinning surgery. Imaging consisted of vascular reactivity measurement upon potent vasodilator administration (Diamox, i.v.) and the dynamic cerebral blood flow parameters assessment using microbubble contrast agent (SovoVue, i.v.). Strikingly, fUS measurements revealed a prominent increase in blood flow and a decrease in vascular reactivity in Dahl-SS rats after 2 weeks of salt diet in cortical, hippocampal and subcortical areas. While the majority of Dahl-SS rats did not show any lesions, hemorrhages or Gd extravasation in MRI, T2 relaxivity across the cortex was lower in Dahl-SS than controls and showed a progressive decrease in the striatum. The state-of-the-art functional imaging readouts enable the detection of vascular dysfunction before development of the structural changes and is a useful tool for investigating the mechanisms of and biomarkers for cerebrovascular disease.

Disclosures: **A. Bialas:** A. Employment/Salary (full or part-time); Sanofi. **R. Immonen:** A. Employment/Salary (full or part-time); Charles River Discovery Services. **J. Uhari-Väänänen:** A. Employment/Salary (full or part-time); Charles River Discovery Services. **H. Vaherto:** A. Employment/Salary (full or part-time); Charles River Discovery Services. **T. Miettinen:** A. Employment/Salary (full or part-time); Charles River Discovery Services. **A. Shatillo:** A. Employment/Salary (full or part-time); Charles River Discovery Services. **J. Dodge:** A. Employment/Salary (full or part-time); Sanofi.

Poster

PSTR130. Vascular Changes in Alzheimer's Disease

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR130.03/E2

Topic: C.02. Alzheimer's Disease and Other Dementias

Support: NIH Grant RF1NS116450
NIH Grant R01AG067018
NIH Grant RF1AG025516

Title: Cerebrovascular tortuosity as a potential early biomarker in Alzheimer’s Disease - optical and MRI study in a mouse model

Authors: *N. SCHWEITZER[1], Y. ZHAO[2], C. COVER[1], M. WU[1], H. AIZENSTEIN[1], A. VAZQUEZ[3], B. IORDANOVA[4];
[1]Univ. of Pittsburgh, PITTSBURGH, PA; [2]Univ. of Pittsburgh, Univ. of Pittsburgh, Oakland, PA; [3]Univ. of Pittsburgh, [4]Univ. of Pittsburgh, Pittsburgh, PA

Abstract: Mounting evidence in the last decade suggests that cerebrovascular dysfunction in Alzheimer’s Disease (AD) emerges decades before cognitive symptoms. We aim to characterize cerebrovascular morphology changes across the lifespan of AD and wild-type (WT) mice in both sexes. A greater understanding of the cerebrovascular morphology changes during dementia etiology and progression can deliver potential biomarker and early treatment targets for AD. We used transgenic AD mouse model (B6C3.Tg.APPswe-PSEN1de9 and age-matched B6C3 wild

type (WT) controls). All vessels were labelled *in vivo* with SR101 and imaged with two-photon microscopy (2PM) at a micron resolution in several cortical areas (n=9 male, 7 female, 3-18 months). The whole brain arterial tree was also imaged with time-of-flight (TOF) MRI (n=23 males, 24 females, 2-25 months). For both TOF and 2PM, 20 images were semi-automatically segmented using Ilastik[1] and manual touch-ups, and then separate 3D 5-layer residual U-Nets were trained on a dataset (600 epochs, Adam optimizer, 1e-4 learning rate, dice loss function). Segmentations were skeletonized, and tortuosity was defined as the arc length divided by Euclidean distance averaged over all branches. For 2PM dataset, junction density was defined as the total number of junctions in a Z-stack divided by the volume of the first 50 images in the stack and then log-transformed. For TOF dataset, images were registered to a B6C3 mouse MRI atlas[2] using AFNI. In the 2PM dataset, we observed small vessels increasing tortuosity in AD mice at 5 months that plateaued after 10 months. In contrast, the WT mice tortuosity did not change until 10 months and then increased during old age. Moreover, AD mice had significantly higher tortuosity than WT mice at age 5-10 months ($p < 1e-7$). We also observed a significant decrease in junction density for AD mice ($p = 0.013$) but not WT mice ($p = 0.25$). Sex differences were not observed for tortuosity, junction density in the 2PM dataset. For TOF MRI dataset that captures mostly larger arteries, we did not observe tortuosity group differences. Instead, we observed significant group and regional differences for the distribution of branch radii and total length of skeleton. Our results suggest that tortuosity of the small vessels, but not the large arteries, occurs early in the disease course of AD. References: 1.) Berg et al (2019) Nature Methods 2.) Ma et al (2005) Neuroscience.

Disclosures: **N. Schweitzer:** None. **Y. Zhao:** None. **C. Cover:** None. **M. Wu:** None. **H. Aizenstein:** None. **A. Vazquez:** None. **B. Iordanova:** None.

Poster

PSTR130. Vascular Changes in Alzheimer's Disease

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR130.04/E3

Topic: C.02. Alzheimer's Disease and Other Dementias

Support: NMRC MOH-OFLCG

Title: Alzheimer's disease patient induced pluripotent stem cell-derived endothelia exhibit defective angiogenesis and accelerated senescence

Authors: *Y.-H. YEN[1], C. CHONG[2], G. CHEW[1], H. WEE[3], C. CHEUNG[3], L. ZENG[4], S.-C. ZHANG[1];
[1]Neurosci. and Behavioral Disorders, Duke-NUS Grad. Med. Sch. Singapore, Singapore, Singapore; [2]Inst. of Chinese Med. Sci., Univ. of Macau, Macau, China; [3]Lee Kong Chian Sch. of Med., Nanyang Technol. Univ., Singapore, Singapore; [4]Natl. Neurosci. Inst., Natl. Neurosci. Inst., Singapore, Singapore

Abstract: Alzheimer's disease (AD) is characterized by an insidious onset of neurocognitive decline, extracellular beta-amyloid (Aβ) plaques, and intracellular tau tangles. Recent studies have highlighted microvascular alterations and transcriptomic changes in endothelial cells (ECs) in post-mortem AD patient brains and AD transgenic mouse models. Microvascular alterations have been observed as early as postnatal day seven in the AD mouse model, suggesting early vascular involvement in AD. However, the specific contribution of ECs to AD in humans and whether these changes are a consequence or a factor in AD development remain unknown. Using induced pluripotent stem cells (iPSCs) derived from familial Alzheimer's disease (FAD) patients with presenilin-1 (PS1) mutations, we demonstrate innate alterations in ECs, including increased proliferation, migration, and tube formation capabilities, as well as reduced barrier integrity in early passages. In later passages, PS1-FAD iPSC-ECs exhibit signs of accelerated cellular senescence and markedly reduced angiogenic properties. Bulk RNA sequencing analysis of the ECs points to potential alterations in the VEGFA-VEGFR2 pathway. Furthermore, deconvolution of our bulk RNA-seq data with existing single-nucleus RNA sequencing data from sporadic AD patient vasculature shows that PS1 FAD-iPSC ECs are transcriptionally more similar to brain endothelial cells of AD patients than age-matched healthy controls. These findings suggest that the vascular changes observed in our PS1-FAD iPSC-EC model may be a shared phenomenon in AD. We propose that dysregulated EC function leads to microvasculature deficits in the brain, contributing to AD pathogenesis.

Disclosures: Y. Yen: None. **C. Chong:** None. **G. Chew:** None. **H. Wee:** None. **C. Cheung:** None. **L. Zeng:** None. **S. Zhang:** None.

Poster

PSTR130. Vascular Changes in Alzheimer's Disease

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR130.05/E4

Topic: C.02. Alzheimer's Disease and Other Dementias

Support: NIH Grant AG066162
University of Toronto Fellowship

Title: Large vessel dilatation and linkage to vascular dementia and Alzheimer's disease pathology

Authors: *D. SIMPSON[1], C. MORRONE[2], D. WEAR[3], H. (. YU[4], J. GUTIERREZ[5]; [2]Work - CAMH, [1]Ctr. for Addiction and Mental Hlth., Toronto, ON, Canada; [3]Home, Univ. of Toronto, Amherstburg, ON, Canada; [4]Ctr. for Addiction and Mental Hlth., Univ. of Toronto, Toronto, ON, Canada; [5]Columbia Univ., New York, NY

Abstract: Extensive post-mortem investigations have yielded compelling evidence pointing towards vascular abnormalities in Alzheimer's disease (AD). While most studies have focused on the association with small blood vessels, a notable minority of AD patients also have

enlargement of larger blood vessels, termed dolichoectasia. Moreover, this particular condition seems to be more prevalent among underrepresented Hispanic, African-American and some Asian populations. In order to effectively address racial and ethnic diversity of AD, it is crucial to gain a comprehensive understanding of the variability in AD pathophysiology across populations. Such knowledge holds the potential to enable researchers in developing enhanced treatments aimed at slowing down or preventing the progression of this disease. Unfortunately, the precise contribution of dolichoectasia to neuroinflammation, proteostasis changes, and AD pathology remains inadequately defined. Thus, acquiring a deeper comprehension will significantly facilitate early detection and treatment, prior to disease progression. To dissect the impact of dolichoectasia in AD, we used several mouse models of Alzheimer's disease (AD; AppNL-F, AppNL-G-F, MAPT, and APPNL-F/MAPT double knock-in) and induced dilatation using 15μU elastase into the cisterna magna. Following dilatation, we tested animals at 3, 6, and 12-month using the Barnes Maze and evidence of AD related pathology and proteostasis changes. Our study using elastase injection showed several lines of compelling evidence linking large vessel dilatation with AD. First, we observed vessel enlargement and tortuosity of major arterial supplies. Second, our findings indicate a trend in Barnes maze measures, suggesting the potential presence of cognitive impairments post-elastase injection. We also noted increased amyloid plaques (ThioS) and tau hyperphosphorylation (PHF1) with dolichoectasia and reduced mature neurons (NeuN). Notably, we detected substantial neuroinflammation (iba1 and GFAP) and increased protein matrix metalloproteinases (MMPs), suspected to play a critical responsive role in dolichoectasia. Finally, we see evidence of impaired proteostasis mechanisms, including increased aggregates (p62). The exciting potential lies in unraveling the intricate connection between dolichoectasia and Alzheimer's disease, opening doors to a wealth of valuable insights. With a deep understanding of this link, we can unlock the secrets of effective biomarkers and targeted therapies in the fight against Alzheimer's.

Disclosures: **D. Simpson:** None. **C. Morrone:** None. **D. Wear:** None. **H.(. Yu:** None. **J. Gutierrez:** None.

Poster

PSTR130. Vascular Changes in Alzheimer's Disease

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR130.06/E5

Topic: C.02. Alzheimer's Disease and Other Dementias

Support: NIH/NIA: 1P01AG052350-06
Alzheimer's Association 008369-00001
NIH/NIA ADRC 5P30AG066530-04

Title: Neurovascular dysfunction predicts white matter connectivity changes underlying cognitive dysfunction in multiple domains in APOE4 carriers.

Authors: *B. ZLOKOVIC[1], A. CHAKHOYAN[1], I. PAPPAS[2], S. BARNES[5], A. SAGARE[1], E. NOUROLLAHIMOGHADAM[1], E. M. REIMAN[6], R. J. CASELLI[7], T. BENZINGER[8], J. C. MORRIS[9], J. M. RINGMAN[10], H. N. YASSINE[11], L. S. SCHNEIDER[3], H. CHUI[4], A. W. TOGA[2], D. A. NATION[12];
[1]Physiol. and Neurosci., Keck Sch. of Med. of USC, Los Angeles, CA; [2]Lab. of Neuro Imaging, USC Stevens Neuroimaging and Informatics Inst., [3]Dept. of Psychiatry and Behavioral Sci., USC, Los Angeles, CA; [4]Dept. of Neurol., USC, Pasadena, CA; [5]Dept. of Radiology, Loma Linda Univ., Loma Linda, CA; [6]Banner Alzheimer Inst., Phoenix, AZ; [7]Mayo Clin., Scottsdale, AZ; [8]Mallinckrodt Inst. of Radiology, Washington Univ. in St. Louis Sch. of Med., St. Louis, MD; [9]Dept. of Neurol., Washington Univ. Sch. of Med., Saint Louis, MO; [10]Dept. of Neurol., [11]Dept. of Med., Keck Sch. of Med., Los Angeles, CA; [12]Dept. of Psychology, Univ. of California Irvine, Irvine, CA

Abstract: Vascular contributions to cognitive impairment in Alzheimer's disease (AD) have been increasingly recognized. *APOE4*, a major susceptibility gene for AD, exerts considerable cerebrovascular toxicity in animal models and in humans. Here, we studied regional cerebral blood flow (CBF) and blood-brain barrier (BBB) changes using multiphase pseudocontinuous spin labeling (pCASL) and dynamic contract enhanced (DCE)-MRI, respectively, in relation to memory and attention/executive function and structural white matter connectivity changes studied by diffusion tensor imaging (DTI)-MRI metrics and DTI-tractography in cognitively unimpaired (CU) *APOE4* carriers (e3/e4, e4,e4; CDR=0; n=50) and *APOE3* homozygotes (CDR=0; n=70) and in *APOE4* carriers (e3/e4, e4,e4; CDR 0.5; n=19) and *APOE3* homozygotes (CDR 0.5; n=24) with clinical decline. We focused on 3 memory regions [hippocampus (HC), parahippocampal gyrus (PHG) and posterior cingulate gyrus (PCG)], and the inferior parietal lobe (IPL), a multimodal sensory area also involved in the attention/executive function. For memory-related connectivity analysis, we studied changes in fractional anisotropy (FA) along the uncinate fasciculus (UF) and cingulum tracts that originate from HC and PCG, respectively, and the superior longitudinal fasciculus (SLF) tract for the attention/executive function. 2x2 ANCOVA (*APOE3* vs *APOE4* carriers x CDR 0 vs 0.5) comparisons were supplemented with hierarchical logistic regression analysis for predicting cognitive impairment. We did not observe changes in CBF in HC or PHG among the 4 groups, while CBF was reduced in PCG in CDR 0.5 vs. CDR 0 *APOE4* carriers. The Ktrans values from DCE-MRI study were significantly higher in HC, PHG and PCG in CU *APOE4* carriers compared to *APOE3* homozygotes which worsened with clinical decline. In *APOE4* carriers BBB disruption in HC, PHG and PCG predicted cognitive impairment (CDR 0.5 vs. CDR 0) and structural changes in the UF and cingulum tracts. CBF was reduced in IPL which predicted impaired attention/executive function in *APOE4* carriers and was associated with disrupted fibers in the SLF tracts. Thus, neurovascular dysfunction predicted structural connectivity changes underlying cognitive dysfunction in multiple domains in *APOE4* carriers.

Disclosures: B. Zlokovic: None. **A. Chakhoyan:** None. **I. Pappas:** None. **S. Barnes:** None. **A. Sagare:** None. **E. Nourollahimoghadam:** None. **E.M. Reiman:** None. **R.J. Caselli:** None. **T. Benzinger:** None. **J.C. Morris:** None. **J.M. Ringman:** None. **H.N. Yassine:** None. **L.S. Schneider:** None. **H. Chui:** None. **A.W. Toga:** None. **D.A. Nation:** None.

Poster

PSTR130. Vascular Changes in Alzheimer's Disease

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR130.07/E6

Topic: C.02. Alzheimer's Disease and Other Dementias

Support: AG052354
AG062738

Title: Diminished pericyte remodeling in the TgSwDI mouse model of cerebral amyloid angiopathy

Authors: *C. D. NIELSON[1], S. K. BONNEY[4], M. J. SOSA[4], A. Y. SHIH[4,2,3];
[1]Grad. Program in Neurosci., [2]Dept. of Pediatrics, [3]Dept. of Bioengineering, Univ. of Washington, Seattle, WA; [4]Ctr. for Developmental Biol. and Regenerative Med., Seattle Children's Res. Inst., Seattle, WA

Abstract: Capillary pericytes serve myriad functions in cerebrovascular regulation and are lost at an accelerated rate in Alzheimer's disease and related dementias. This loss is correlated with blood-brain barrier damage, neuron death, and increased amyloid burden. Recently, it has also been demonstrated that pericyte loss contributes to changes in capillary flow and structure. Optical ablation of brain pericytes leads to the dilation of uncovered vessels, creating a maldistribution of blood flow within the capillary bed and increasing the incidence of capillary stalls and regressions. However, pericytes possess an intrinsic repair strategy to compensate for pericyte loss. The surviving pericytes remodel their processes to restore pericyte coverage of the exposed endothelium. Pericyte remodeling is impaired in the aged mouse brain, but we still have limited knowledge of how pericyte remodeling is affected by Alzheimer's disease. To investigate this question, TgSwDI mice that express the Swedish, Dutch, and Iowa mutations of the *APP* gene were bred to PDGFRβCre;Ai14 mice that express tdTomato in all mural cells. Chronic cranial windows were implanted in mice aged 12-18 months, *in vivo* optical ablation of brain pericytes was performed, and pericyte remodeling was measured over the span of one week (n = 3 mice/group). Pericyte process growth was significantly reduced in TgSwDI mice compared to age-matched littermate controls, with growth most dramatically reduced in the postcapillary zone. Additionally, pericyte processes in TgSwDI mice occasionally retracted rather than grew, increasing the amount of uncovered vascular length. Finally, regions of vasculature uncovered by pericytes were often present in TgSwDI mice prior to pericyte ablation, with loss most often observed on postcapillary venules. Our results suggest that Alzheimer's disease may exacerbate the pericyte remodeling deficits that are seen during aging and that pericytes on the venular side of the microvascular network may be particularly vulnerable to disease. Ongoing work is aimed towards dissecting mechanisms of pericyte structural plasticity and therapeutic strategies for augmentation of pericyte coverage.

Disclosures: C.D. Nielson: None. **S.K. Bonney:** None. **M.J. Sosa:** None. **A.Y. Shih:** None.

Poster

PSTR130. Vascular Changes in Alzheimer's Disease

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR130.08/E7

Topic: C.02. Alzheimer's Disease and Other Dementias

Title: Blood-brain-barrier microphysiological models for increased predictability of antibody-triggered receptor-mediated transcytosis.

Authors: ***G. FEWELL**[1,2,3], J. ROSANO[1], B. LI[2], C. SODJA[2], C. CHARLEBOIS[2], Q. LIU[2], D. STANIMIROVIC[2], D. RAMSEY[1], A. JEZIERSKI[2];
[1]SynVivo Inc., Huntsville, AL; [2]Translational Biosci., Human Hlth. Therapeut. Res. Center, Natl. Res. Council of Canada, Ottawa, ON, Canada; [3]Univ. of Ottowa, Ottawa, ON, Canada

Abstract: Microphysiological systems (MPS) that model the blood brain barrier (BBB) *in vitro* are crucial tools to aid in the pre-clinical evaluation and selection of BBB-permeant biotherapeutics. BBB models can be created using cells from various sources, including cell lines, primary cells, or iPSC-derived cells. Factors such as fluid shear stress, as well as co-culture alongside astrocytes and pericytes, have been shown to affect barrier integrity and the permeability of drugs across the BBB. To date, there are no studies directly comparing the choice of cell sources in flow-based human BBB tri-culture models. We developed human BBB-on-chip models using the SynVivo microfluidic platform (SynBBB) and a variety of endothelial cell sources, including iPSC-derived brain endothelial-like cells (iBECs), hCMEC/D3 cell line, and primary brain endothelial cells. Brain microvessel lumens were established under physiological shear stress conditions to induce tight barrier formation, resulting in a measurable decrease in paracellular permeability to dextran and sodium fluorescein under flow conditions, as well as in neurovascular tri-cultures containing astrocytes and pericytes. Similar trends were observed using real-time on-chip trans-endothelial electrical resistance (TEER) measurements. For the iBEC-based SynBBB model, shear stress was also found to induce endothelial cell maturation. We deployed these SynBBB tri-culture models to study antibody-triggered receptor mediated transcytosis by perfusing the brain endothelial lumens with a well-characterized single domain BBB-carrier, FC5-Fc, and non-crossing A20.1 control. We observed a significant increase in FC5-Fc transcytosis under physiological shear stress conditions compared to static cultures. Collectively, these findings suggest that SynBBB technology can recapitulate the physiological characteristics of the BBB *in vivo* and offer a more predictive platform for assessing antibody transcytosis across the BBB.

Disclosures: **G. Fewell:** A. Employment/Salary (full or part-time); SynVivo Inc. **J. Rosano:** A. Employment/Salary (full or part-time); SynVivo Inc. **B. Li:** A. Employment/Salary (full or part-time); National Research Council of Canada. **C. Sodja:** A. Employment/Salary (full or part-time); National Research Council of Canada. **C. Charlebois:** A. Employment/Salary (full or part-time); National Research Council of Canada. **Q. Liu:** A. Employment/Salary (full or part-time); National Research Council of Canada. **D. Stanimirovic:** A. Employment/Salary (full or part-time); National Research Council of Canada. **D. Ramsey:** A. Employment/Salary (full or part-time); SynVivo Inc. **A. Jezierski:** A. Employment/Salary (full or part-time); National Research Council.

Poster

PSTR130. Vascular Changes in Alzheimer's Disease

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR130.09/E8

Topic: C.02. Alzheimer's Disease and Other Dementias

Support: NIA R01AG065836

Title: Vascularamyloidbload in the meningeal arterial network correlates with loss of cerebral blood flow and pial collateral vessel remodeling in a murine model of Alzheimer's disease

Authors: A. KALOSS[1], J. LI[2,3], ***J. BROWNING**[4], Y. PAN[2], H. SONTHEIMER[5], M. THEUS[1,2], M. L. OLSEN[2];
[1]Dept. of Biomed. Sci. and Pathobiology, [2]Sch. of Neurosci., [3]Genetics, Bioinformatics and Computat. Biol., [4]Virginia Tech., Blacksburg, VA; [5]Dept. of Neurosci., Univ. of Virginia, Charlottesville, VA

Abstract: Decades of research indicate that a global reduction in cerebral blood flow (CBF) represents one of the earliest observable pathologies in individuals with Alzheimer disease (AD), a phenotype that is recapitulated in animal models of AD with Aβ accumulation. Reduced CBF occurs prior to significant parenchymal plaque accumulation or neurological decline and is now considered predictive and a potential diagnostic indicator for early-stage AD. Chronic reduction in CBF and the subsequent reduction in parenchymal tissue oxygenation and glucose may be sufficient to drive neurodegeneration, yet, the underlying cause of global reduced CBF is not understood. Using methoxy-X04 to label vascular Aβ, and vascular labeling approaches, we identified significant Aβ burden on the pial artery/arteriole network in J20 (PDGF-APPSwInd) AD mice. *Imaris* image analysis of Aβ accumulation on reconstructed vessels indicated a progressive increase in Aβ burden on both the MCA and ACA, reaching 40% coverage in mice one year of age. No sex differences were observed. Additionally, the leptomeningeal anastomoses or pial arteriole collateral vessels, i.e. the by-pass system responsible for re-routing blood flow in the event of an occlusion, displayed active outgrowth and remodeling, including an increase in MCA-ACA collateral diameter and collateral tortuosity in J20 male and female mice by one year of age. Despite increased collateral diameter, overall surface hemisphere perfusion measured by laser speckle imaging indicate CBF was decreased by 15% in 12-month J20 mice compared to age matched WT littermates. Our findings indicate significant Aβ burden on the meningeal arteries, the inflow for parenchymal perfusion, may serve to restrict global CBF observed in animal models of AD. Further we have identified retrograde reperfusion initiated through collateral vessel remodeling as a potential compensatory mechanism to restore CBF to affected brain tissue.

Disclosures: A. Kaloss: None. **J. Li:** None. **J. Browning:** None. **Y. Pan:** None. **H. Sontheimer:** None. **M. Theus:** None. **M.L. Olsen:** None.

Poster

PSTR130. Vascular Changes in Alzheimer's Disease

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR130.10/E9

Topic: C.02. Alzheimer's Disease and Other Dementias

Support: NIH Grant P01AG052350
NIH Grant P30AG06653
Cure Alzheimer's Fund
Alzheimer's Association strategic grant 509279
the Foundation Leducq Transatlantic Network of Excellence for the study of Perivascular Spaces in Small Vessel Disease reference no. 16 CVD 05
Open Philanthropy to B.V.Z.

Title: Biofluid biomarkers of brain vascular dysfunction and Alzheimer's disease in APOE4 carriers

Authors: *A. P. SAGARE[1], D. A. NATION[5], I. RAMIREZ[1], C. TORRES-SEPULVEDA[1], E. B. JOE[2], H. C. CHUI[2], J. M. RINGMAN[2], M. G. HARRINGTON[2], H. N. YASSINE[2], L. S. SCHNEIDER[2], K. M. KIRMESS[6], M. R. MEYER[6], K. YARASHESKI[6], A. W. TOGA[3], B. V. ZLOKOVIC[4];
[1]Physiol. and Neurosci., [2]Alzheimer's Dis. Res. Center, Dept. of Neurology, Keck Sch. of Med., [3]Inst. for Neuroimaging and Informatics, Keck Sch. of Med., [4]Physiol. and Neuroscience, Zilkha Neurogenetic Inst., USC, Los Angeles, CA; [5]Inst. for Memory Disorders and Neurolog. Impairments, Univ. of California, Irvine, Irvine, CA; [6]C2N Diagnostics, St. Louis, MO

Abstract: Recent studies have shown that brain vascular dysfunction and blood-brain barrier (BBB) disruption contribute to cognitive decline and Alzheimer's disease (AD). With the advancements in neuroimaging techniques and the development of high-sensitivity methods for the measurement of AD biomarkers such as amyloid-β (Aβ) and p-tau and vascular injury biomarkers in cerebrospinal fluid (CSF) and blood, it is now feasible to assess the impact of vascular dysfunction in the pathophysiology of AD, including the effect of *APOE4*, a significant risk factor for AD. Our earlier work has shown that CSF biomarkers of neurovascular dysfunction are elevated in the early stages of cognitive impairment, especially in *APOE4* carriers. We also found that soluble platelet-derived growth factor-β (sPDGFRβ), a marker associated with brain microvascular pericyte damage, may be used as a predictive biomarker of future cognitive decline in *APOE4* carriers. Due to their practical advantages, there is a growing interest in identifying blood-based biomarkers that could be used for diagnostic purposes in clinics and clinical trials. Here, we quantified baseline plasma levels of Aβ42/40 in 162 plasma samples obtained from human research participants ≥45 years of age recruited at the University of Southern California Alzheimer's Disease Research Center (ADRC) using C_2N Diagnostics' immunoprecipitation liquid chromatography-tandem mass spectrometry-analytical method (PrecivityAD®). sPDGFRβ levels were analyzed using an ELISA assay that we optimized for

plasma. We stratified participants by *APOE* genotype as *APOE4* carriers (Ɛ3/Ɛ4 and Ɛ4/Ɛ4, *n*=66) or *APOE4* non-carriers (Ɛ3/Ɛ3, *n*=96). We found that plasma Aβ42 levels and Aβ42/40 ratio were significantly lower in *APOE4* carriers compared to *APOE3* carriers ($p<0.01$). Additionally, the Amyloid Probability Score (APS), which reflects the likelihood of cerebral amyloid pathology, was significantly higher in *APOE4* carriers compared to *APOE3* carriers ($p<0.0001$). Plasma Aβ42 levels and the Aβ42/40 ratio correlated positively with CSF Aβ42 levels ($p<0.01$) and negatively with APS ($p<0.001$). We found higher plasma sPDGFRβ levels in *APOE4* carriers compared to *APOE3* carriers ($p<0.05$). In this longitudinal study cohort, we aim to further assess the utility of plasma amyloid and vascular injury biomarkers for reliably identifying individuals at risk of future cognitive decline.

Disclosures: **A.P. Sagare:** None. **D.A. Nation:** None. **I. Ramirez:** None. **C. Torres-Sepulveda:** None. **E.B. Joe:** None. **H.C. Chui:** None. **J.M. Ringman:** None. **M.G. Harrington:** None. **H.N. Yassine:** None. **L.S. Schneider:** None. **K.M. Kirmess:** None. **M.R. Meyer:** None. **K. Yarasheski:** None. **A.W. Toga:** None. **B.V. Zlokovic:** None.

Poster

PSTR130. Vascular Changes in Alzheimer's Disease

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR130.11/E10

Topic: C.02. Alzheimer's Disease and Other Dementias

Support: Institutional Development Award from the National Institutes of Medical Sciences of the National Institutes of Health Grant # 3P2012130701A1S1
Institutional Development Award from the National Institutes of Medical Sciences of the National Institutes of Health Grant # HL149264-01A1

Title: Increased Plasma Cystathionine Beta-Synthase in Alzheimer's Disease and Related Dementias (ADRD): Inverse Correlation with Cognitive Performance.

Authors: *K. T. LOFTON[1], T. REEKES[5], A. ALHAQUE[2], E. A. DISBROW[1], K. Y. STOKES[2], J. C. PATTERSON, II[3], C. G. KEVIL[4], J. S. ALEXANDER[2];
[1]Ctr. for Brain Hlth., [2]Dept. of Mol. and Cell. Physiol., [3]Dept. of Psychiatry and Behavioral Med., [4]Dept. of Pathology, LSUHS, Shreveport, LA; [5]Dept. of Anesthesiol., Duke Univ. Med. Ctr., Durham, NC

Abstract: The "*vascular hypothesis*" of Alzheimer's disease (AD) states that neurodegenerative proteinopathy is preceded by cerebral hypoperfusion and brain microvascular abnormalities. Hydrogen sulfide and its metabolites are important in neuromodulation and vasoregulation, and we have described a link between redox-related *disturbances in sulfide metabolism* and measures of cerebrovascular disease, brain atrophy and cognitive disturbance in human AD and related dementias (ADRD). However, the tissue and enzymatic sources of these elevated sulfides still remain unclear. Using immunoblotting, we compared levels of 3 enzymatic sources of plasma

sulfides, cystathionine beta synthase (CBS), cystathionine gamma-lyase (CSE), 3-mercaptopyruvate sulfurtransferase (3-MST) as well as plasma beta-amyloid. Neuropsychological test results, demographic data, and whole blood samples were collected from healthy controls (N = 27; mean age 71.3 (SD = 6.3) years) and individuals who met the criteria for ADRD (AD Dementia Assessment Scale-cognitive subscale score >=17; N = 37; mean age 74.3 (SD = 6.80) years). Plasma (0.5ul) CBS, MST, CSE or beta amyloid was immunoblotted and imaged using HRP/ECL reagent on a Chemidoc Image Analyzer. Plasma levels of free, acid-labile, bound, and total hydrogen sulfide were evaluated using the monobromobimane method. Plasma CBS was increased in ADRD (2.43-fold higher, $p < 0.001$) vs. controls. CSE and MST were not significantly different between groups. Plasma beta amyloid was also significantly increased in ADRD (19%, p=0.0121). CBS was positively associated with poorer ADAS-Cog scores (R=0.455, p<0.0001) while CSE and MST were not. Plasma beta amyloid was significantly correlated with acid-labile sulfide ($R = 0.265$, $p = 0.041$), plasma CBS ($R = 0.254$, $p = 0.043$), and plasma MST ($R = 0.619$, $p < 0.001$). While cellular sources of these sulfides remains to be determined, the cellular source of CBS may be endothelial cells which lie at the interface of the brain and blood and express CBS. A better understanding of the underlying vascular pathomechanisms and consequences that confer greater risk due to excess sulfide burden may lead to improved detection and treatment of ADRD.

Disclosures: **K.T. Lofton:** None. **T. Reekes:** None. **A. Alhaque:** None. **E.A. Disbrow:** None. **K.Y. Stokes:** None. **J.C. Patterson:** None. **C.G. Kevil:** None. **J.S. Alexander:** None.

Poster

PSTR130. Vascular Changes in Alzheimer's Disease

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR130.12/E11

Topic: C.02. Alzheimer's Disease and Other Dementias

Support: Institutional Development Award from The National Institutes of Medical Sciences of the National Institutes of Health Grant # 3P2012130701A1S1
Institutional Development Award from The National Institutes of Medical Sciences of the National Institutes of Health Grant #HL149264-01A1

Title: Alzheimer's Disease and the Cerebellum: Correlation of Circulating Plasma Sulfide Levels with Cerebellar Atrophy and Cognitive Impairment

Authors: *P. M. LUTHER[1], K. G. YABUT[1], K. LOFTON[2], T. H. REEKES[7], C. R. LEDBETTER[3], K. Y. STOKES[4], J. S. ALEXANDER[4], C. G. KEVIL[5], J. C. PATTERSON, II[6], E. A. DISBROW[2];
[1]Louisiana State Univ. Hlth. Sci. Ctr. at Shreveport, Sch. of Med., Shreveport, LA; [2]Ctr. for Brain Hlth., [3]Dept. of Neurosurg., [4]Dept. of Mol. and Cell. Physiol., [5]Dept. of Pathology, [6]Dept. of Psychiatry and Behavioral Med., Louisiana State Univ. Hlth. Sci. Ctr. at Shreveport, Shreveport, LA; [7]Dept. of Anesthesiol., Duke Univ. Med. Ctr., Durham, NC

Abstract: Alzheimer's disease and related dementias (ADRD) is associated with loss of cerebellar volume via accumulation of amyloid beta plaques and loss of purkinje cells. The cerebellum subserves cognitive skills such as memory, language, and executive function, and cerebellar dysfunction may be associated with cognitive decline. The vascular contribution to ADRD is well described, and our work links elevated hydrogen sulfide, a vasodilator and neuromodulator, to cognitive dysfunction, brain atrophy and microvascular disease in human ADRD. Our aim was to evaluate the association between sulfide dysregulation and cerebellar atrophy and their relationship to cognitive deficits in ADRD.We collected neuropsychological, plasma sulfide, and cerebellar volume data from 58 participants (29 AD and related dementias or ADRD). We evaluated memory, language and praxis (AD Assessment Scale-Cognitive Subscale (ADAS-Cog), executive function (Trails B-A, D-KEFS), and processing speed (Symbol Digit Modalities Test (SDMT)). 3 plasma sulfide pools were measured: free, acid-labile (e.g.iron-sulfur clusters) and bound (per- and polysulfides) using monobromobimane and 3T MRI normalized cerebellar white and gray matter volume using FreeSurfer 7.2; analysis using IBM SPSS 28.The ADRD group had poorer cognitive performance vs. controls across all measures (ADRD mean (SD) vs Control): ADAS ($F(4,52) = 73.47$, $p < 0.05$), Trails ($F(4,38) = 4.84$, $p < 0.05$), SDMT ($F(4,49) = 18.47$, $p < 0.05$), letter fluency ($F(4,39) = 3.42$, $p = 0.07$), and category switch ($F(4,42) = 12.15$, $p < 0.05$). All three sulfide pools were increased in ADRD subjects including: acid labile ($F(4,52) = 10.44$, $p < 0.05$), bound ($F(4,52) = 4.65$, $p < 0.05$), and total sulfides ($F(4,52) = 17.36$, $p < 0.05$), compared to controls. We found that compared to control subjects, individuals with ADRD had decreased normalized cerebellar white matter volume ($F(4,54) = 3.86$, $p = 0.055$) and cerebellar gray matter ($F=(4,54) = 4.20$, $p < 0.05$). Cerebellar volume measures correlated with plasma sulfides (total sulfides (white matter $R=-.227$, $p<0.05$) and cognition (ADAS (gray matter $R=-.444$,$p<0.05$; white matter $R=-.442$, $p<0.05$), Trails B-A (gray matter $R=-.366$,$p<0.05$; white matter $R=-.381$, $p<0.05$), SDMT (gray matter $R=.402$,$p<0.05$; white matter $R=.397$, $p<0.05$), letter fluency (gray matter $R=.290$,$p<0.05$; white matter $R=.289$, $p<0.05$), and category switch (gray matter $R=.293$,$p<0.05$). Our research indicates for the first time, a significant association between cognitive dysfunction, cerebellar atrophy, and sulfide dysregulation in Alzheimer's Disease and Related Disorders

Disclosures: P.M. Luther: None. **K.G. Yabut:** None. **K. Lofton:** None. **T.H. Reekes:** None. **C.R. Ledbetter:** None. **K.Y. Stokes:** None. **J.S. Alexander:** None. **C.G. Kevil:** None. **J.C. Patterson:** None. **E.A. Disbrow:** None.

Poster

PSTR130. Vascular Changes in Alzheimer's Disease

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR130.13/E12

Topic: C.02. Alzheimer's Disease and Other Dementias

Support: LSU Health Shreveport Institutional Research Funds (CIRP)

Title: Care navigation and research participation of underserved populations with Alzheimer's disease and related dementias

Authors: *F. THOMAS-DEAN[1], T. REEKES[3], C. ARNOLD[2], E. DISBROW[1];
[1]Ctr. for Brain Hlth., [2]Dept. of Intrnl. Med., LSU Hlth. Sci. Center, Shreveport, Shreveport, LA;
[3]Dept. of Anesthesiol., Duke Univ. Med. Ctr., Durham, NC

Abstract: Minorities and people with low income are underrepresented in research on Alzheimer's disease and related dementias (ADRD). Racial diversity in clinical research is essential among patients with ADRD to ensure generalizability of results and to decrease disease burden for Black and African Americans who are disproportionately affected by the disease. Black adults are up to twice as likely as white adults to develop Alzheimer's disease. Yet only 2.4% of participants in randomized AD clinical trials are Black, despite willingness to participate. Barriers to participation include lack of awareness of available clinical trials for ADRD, limited healthcare access, lack of knowledge about ADRD, insufficient community engagement, communication obstacles, distrust of research and researchers and participant and caregiver burden. These problems are amplified in Louisiana which has long standing health and social disparities linked to race, poverty, and rural residence. To address barriers to research participation we evaluated a modified care navigation program as a tool to support participation. Inclusion criteria for patient/caregiver dyads included: ≥55 years of age; had either a physician-based dementia diagnosis or an Alzheimer's Disease Assessment Scale - Cognitive Subscale (ADAS-cog) score ≥ 17; lived in the community. The Care Ecosystem intervention consisted of initial evaluation (Zarit-12 for caregiver burden, Neuropsychiatric Inventory (NPI-Q) for psychiatric and behavioral concerns and ADAS-cog for cognitive function) and a structured monthly follow-up phone interview for 12 months assessing current needs and care-based solutions. 18 dyads were enrolled in the study. 16 dyads completed the care navigation program and 2 withdrew due to family dysfunction and admittance into a nursing home. 6 (37.5%) of the 16 dyads identified as Black and 10 (62.5%) as White. 31.3% reported highest level of education completed was high school or less. 37.5% reported annual income <$45,000. 5 participants were identified as probable ADRD through the care navigation process and referred for appropriate medical care. Regarding dementia disease severity, neuropsychiatric symptoms, or caregiver burden there was no differences between white adults and black adults. Compared to the national average of 2.4% of black participants in ADRD clinical trials, the retention rate in this study was 37.5% among black participants. Our data suggests that care navigation is an effective tool for research recruitment and participation among diverse, underserved, and low-income populations. 15 of the original 18 dyads participated in a separate research study on ADRD.

Disclosures: F. Thomas-Dean: None. **T. Reekes:** None. **C. Arnold:** None. **E. Disbrow:** None.

Poster

PSTR131. Altered Energy Homeostasis in Neurodegenerative Disorders

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR131.01/E13

Topic: C.02. Alzheimer's Disease and Other Dementias

Title: Alterations in sleep, neuronal activity, and metabolism in the P301S PS19 mouse model of tauopathy

Authors: *R. IRMEN[1], C. M. CARROLL[2], N. CONSTANTINO[2], J. A. SNIPES[3], S. L. MACAULEY[1];
[1]Univ. of Kentucky, Lexington, KY; [2]Wake Forest Sch. of Med., [3]Wake Forest Sch. of Med., Winston Salem, NC

Abstract: The two pathological biomarkers for Alzheimer's disease (AD) are amyloid-beta (Aβ) and tau aggregation. While the presence of both biomarkers is necessary for diagnosis, tau pathology coincides with cognitive impairment (CI) and neurodegeneration. Along with CI, AD patients often exhibit symptoms like metabolic and sleep impairments. Currently, it is unclear if symptoms are attributed to Aβ or tauopathy. Previous research using mouse models overexpressing Aβ reveals relationships between Aβ, metabolic, sleep dysfunction. Further, our lab previously demonstrated links between central and peripheral metabolic changes. This bidirectional relationship is well described in mouse models of Aβ overexpression; however, it is unclear in mouse models of tauopathy. Therefore, we investigated how AD-related tau pathology alters sleep, metabolism, and neuronal activity. To explore changes in peripheral metabolism, body weights were measured and glucose tolerance tests were conducted on P301S PS19 and wild type (WT) female mice (n=6-10). Next, the mice were dosed with 2g/kg of glucose followed by blood glucose measurements in 15-minute increments for two hours. The TSE Phenomaster metabolic screening platform tracks indirect calorimetry, food intake, body weight, and activity over three days. Paired glucose and lactate biosensors placed within the mouse hippocampus tracked second by second metabolic fluctuations over three days. EEG and EMG electrodes were placed to record sleep-wake cycles during this period. All methods completed on 3-, 6-, and 9-month-old P301S and WT female mice. As early as six months old, P301S mice exhibit altered sleep/wake activity where P301S mice spend increased time in wake and decreased time in NREM and REM specifically in the light or inactive period. Further, EEG frequency bands, delta, theta, and beta, are altered during various sleep/wake states in the presence of pathology at 6 or 9 months. Suggesting that neuronal activity and sleep are altered as tau pathology develops. Further, tau pathology increases the rate glucose is consumed from the periphery. Data from the TSE suggests that tau pathology differentially alters other peripheral metabolic characteristics. Additionally, when compared to WT mice, P301S mice exhibit altered central metabolic fluctuations where P310S mice conserve glucose and lactate rhythms from 6 to 9 months. The results from this study suggest that tau pathology alters neuronal activity and sleep/wake cycles which therefore disrupts peripheral and central metabolism. Our current research is valuable in expanding the understanding of the impacts that tau pathology has on symptoms involved in AD.

Disclosures: R. Irmen: None. **C.M. Carroll:** None. **N. Constantino:** None. **J.A. Snipes:** None. **S.L. Macauley:** None.

Poster

PSTR131. Altered Energy Homeostasis in Neurodegenerative Disorders

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR131.02/E14

Topic: C.02. Alzheimer's Disease and Other Dementias

Support: UNCF/Bristol-Myers Squibb E.E. Just Faculty Fund
BWF Career Awards at the Scientific Interface Award
BWF Ad-hoc Award
NaNIH Small Research Pilot Subaward to 5R25HL106365-12 from the NIH PRIDE Program, DK020593
Vanderbilt Diabetes and Research Training Center for DRTC Alzheimer's Disease Pilot & Feasibility Program
CZI Science Diversity Leadership grant number 2022- 253529 from the Chan Zuckerberg Initiative DAF

Title: MICOS complex and mitochondria morphology changes in 3D reconstructions of the amygdala and hypothalamus in Alzheimer's disease

Authors: *B. SHAO[1], A. G. MARSHALL[1], C. PALAVICINO-MAGGIO[2,3], A. O. HINTON, Jr.[1];
[1]Dept. of Mol. Physiol. and Biophysics, Vanderbilt Univ., Nashville, TN; [2]Basic Neurosci. Div., McLean Hosp., Belmont, MA; [3]Dept. of Psychiatry, Harvard Med. Sch., Boston, MA

Abstract: Alzheimer's disease (AD) is a neurodegenerative disease distinguished by cognitive and motor impairments associated with mitochondrial dysfunction, altered oxidative phosphorylation, calcium dysregulation, impaired glucose metabolism, and increased apoptosis. The mitochondrial contact site and cristae organizing system (MICOS) complex, particularly the CHCHD6 protein, have been implicated in regulating mitochondrial structure and pathology in AD. However, the impact of MICOS and mitochondrial morphology on neuronal function in regions like the amygdala and hypothalamus remains unclear. Using histology staining to confirm abnormal protein accumulation, we examined postmortem brain samples from neurotypical and AD patients. MICOS gene expression analysis revealed Mic10 downregulation across all AD brain regions and specific downregulation of Mic27 in males with AD. In an analysis of laboratory evolved *Drosophila melanogaster* populations, we observed the downregulation of CHCHD3/6 and the upregulation of mitofilin in flies with advanced aging. Next, we hypothesized that morphology would be altered in AD, mouse-aged samples, and drosophila melanogaster populations. Thus, we did a TEM analysis and showed reduced mitochondrial morphology in AD samples compared to neurotypical ones. Furthermore, using SBF-SEM and 3D reconstruction by Amira software analysis, we investigated mitochondrial morphology in the amygdala and hypothalamus of mice. We found that mitochondrial length, perimeter, 3D area, and volume increased with aging in the amygdala but not in the hypothalamus, while sphericity increased in both regions. Future studies should expand 3D reconstruction to include other neuronal structures, examine MICOS protein expression in human AD neurons, and assess β-amyloid and tau protein expression in MICOS knockouts.

These efforts will provide further insights into the role of MICOS in mitochondrial structure in AD patients' brains.

Disclosures: B. Shao: None. **A.G. Marshall:** None. **C. Palavicino-Maggio:** None. **A.O. Hinton:** None.

Poster

PSTR131. Altered Energy Homeostasis in Neurodegenerative Disorders

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR131.03/E15

Topic: C.02. Alzheimer's Disease and Other Dementias

Support: National Institute on Aging AG065628

Title: Elevated ER-Ca^{2+}signaling in human AD neurons drives disruptions in mitochondrial functions

Authors: *W. GALLEGOS[1], S. MUSTALY-KALIMI[1], I. SEKLER[2], R. A. MARR[1], G. E. STUTZMANN[1];
[1]Neurosci., Rosalind Franklin Univ. of Med. and Sci., North Chicago, IL; [2]Physiol. and Cell Biol., Ben-Gurion Univ. of the Negev, Beer Sheva, Israel

Abstract: The tight coupling between the endoplasmic reticulum (ER) Ca^{2+} channels and the mitochondria is essential for neuronal bioenergetics and cellular metabolism. The Ca^{2+} released from the ER-localized ryanodine receptor (RyR) and inositol triphosphate receptor (IP_3R) channels is taken up by the mitochondria and serves a key regulatory role in oxidative phosphorylation and ATP production. In AD, RyR-evoked Ca^{2+} release is markedly increased and negatively alters a host of downstream physiological functions, including synaptic deficits, protein mishandling, and memory encoding. Here we hypothesize that the dysregulated RyR-Ca^{2+} release from the ER directly alters mitochondrial activity and contributes to increased oxidative stress, impaired bioenergetics, and disrupted ETC functions, and thus is a central feature in AD-related neurodegeneration.To test this, we utilized a series of cellular models, including RyR2-expressing HEK cells and human-induced neurons derived from familial AD and NonAD patients. We monitored mitochondrial functions using live cell fluorescent imaging based on a CCD sCMOS pCO-Edge camera system and Nikon NIS-Elements AR software. Biosensors such as genetically-encoded Ca^{2+} indicators and organelle-targeted fluorophores were used to measure and compare mitochondrial Ca^{2+} uptake, mitochondrial membrane potential, and superoxide production between the AD and NonAD neurons. In neurons from AD patients, resting mitochondrial Ca^{2+} levels, membrane potential, and superoxide production were significantly higher than in non-AD neurons and was normalized by the RyR negative allosteric modulator, Ryanodex (10uM). Furthermore, evoked RyR-Ca^{2+} release generated by 10mM caffeine bath application resulted in significantly elevated mitochondrial Ca^{2+} uptake, depolarization of the mitochondrial membrane potential, and elevated superoxide production

relative to NonAD neurons, and was corrected to the NonAD phenotype with Ryanodex (10uM). The data demonstrate that elevated RyR-Ca^{2+} signaling elicits a maladaptive response within the mitochondria that links with AD features such as impaired cellular metabolism and increased free radical production. Thus, identifying the pathogenic cascades within the mitochondria that result from elevated ER-Ca^{2+} release is essential for elucidating cellular disease mechanisms in early AD.

Disclosures: W. Gallegos: None. **S. Mustaly-Kalimi:** None. **I. Sekler:** None. **R.A. Marr:** None. **G.E. Stutzmann:** None.

Poster

PSTR131. Altered Energy Homeostasis in Neurodegenerative Disorders

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR131.04/E16

Topic: C.02. Alzheimer's Disease and Other Dementias

Support: NIA 1R01AG062762-01A1
NIA 5R01AG062762-02
NIH 1R56AG062762-01

Title: Exacerbation of beta-amyloid and diabetes-associated pathology due to chronic hyperglycemia in an Alzheimer's disease mouse model

Authors: *A. A. ORTIZ, A. M. OSSE, J. WANG, B. BALSAMO, L. B. PASIA, K. D. SUK, K. S. HERNANDEZ, L. CORTEZ, J. W. KINNEY;
Brain Hlth., Univ. of Nevada Las Vegas, Las Vegas, NV

Abstract: Alzheimer’s disease (AD) is a neurodegenerative disease that is characterized by progressive synaptic and neuronal loss, learning and memory deficits, and cognitive decline. AD affects over 6.5 million Americans and is the 6th leading cause of death in the US. Furthermore, by 2050, the US is projected to spend over $1.1 trillion on AD-related treatments.Pathological hallmarks of AD include senile beta-amyloid (Aβ) plaques, intracellular neurofibrillary tangles (NFTs), and chronic neuroinflammation, which can promote and exacerbate both Aβ and NFTs levels. AD is classified as early onset (EOAD) or late onset (LOAD). EOAD is associated with genetic mutations and accounts for 3-5% of all AD cases. In contrast, LOAD accounts for 95-97% of all AD cases with no genetic etiology; however, several genetic and/or other comorbidities confer increased risk for LOAD. Diabetes mellitus (DM) is a major risk factor for AD. DM confers up to a 4-fold increased risk for developing AD, and approximately 81% of individuals with AD have type II diabetes or are glucose intolerant. Hyperglycemia – abnormal elevated blood glucose levels – is the primary characteristic of DM. We have previously shown that chronic hyperglycemia can initiate and promote neuroinflammation, resulting in significant increases in hyperphosphorylated tau protein (pTau), learning and memory impairments, and other AD-related targets that are consistent with other AD models. However, the role of chronic

hyperglycemia and its impact on Aβ are still being elucidated.Alterations in γ-aminobutyric acid (GABAB), the brain's primary inhibitory neurotransmitter, have been implicated in AD. GABA (B) receptors are expressed on glia cells, and play a role in neuroinflammation. In this experiment, we induced a chronic hyperglycemic state in a GABA (B) receptor knockdown mouse model restricted to microglia (Osse, 2023), crossed with the APP/PS1 mouse model, that results in increased amyloid processing. We administered low and staggered dosages of streptozotocin (STZ) over a period of 6 weeks to induce hyperglycemia; glucose levels were taken weekly. The aim was to investigate the additive effects of elevated glucose on Aβ load and other AD-DM-related markers.Briefly, our data indicate altered fasting blood glucose levels, Aβ load, and other AD-DM-related targets in the hippocampus; a region that is first affected by AD. These findings suggest that chronic hyperglycemia can exacerbate the accumulation of Aβ plaques and contribute to the dysregulation of AD-DM-related markers. Further investigation into the mechanisms underlying these interactions may provide valuable insights into the link between DM and AD.

Disclosures: A.A. Ortiz: None. **A.M. Osse:** None. **J. Wang:** None. **B. Balsamo:** None. **L.B. Pasia:** None. **K.D. Suk:** None. **K.S. Hernandez:** None. **L. Cortez:** None. **J.W. Kinney:** None.

Poster

PSTR131. Altered Energy Homeostasis in Neurodegenerative Disorders

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR131.05/E17

Topic: C.02. Alzheimer's Disease and Other Dementias

Title: Roles of IGF1 in hippocampus and adipose tissue in APP/PS2 transgenic mouse

Authors: D. JO[1], ***J. SONG**[2];
[1]Dept. of anatomy, Chonnam national university medical shool, Hwasun, Korea, Republic of;
[2]Dept. of anatomy, Chonnam Natl. Univ. Med. Sch., Hwasun, Korea, Republic of

Abstract: Roles of IGF-1 in hippocampus and adipose tissue in APP/PS2 transgenic mouse
Danbi Jo[1], Juhyun Song[1+]
[1]Chonnam national university medical school, Department of anatomy
AbstractAlzheimer's disease (AD) is the most common neurodegenerative disease and presents various pathological changes including high amyloid beta aggregation, severe neuroinflammation, and neuroglial activation. Given previous studies showing that excessive adiposity in patient with metabolic dysfunction are associated with memory impairment, we investigated genetic changes both in the hippocampus and visceral adipose tissues in APP/PS2 transgenic mice as AD mouse model. Insulin like growth factor-1 (IGF-1) is known to regulate insulin sensitivity, glucose homeostasis, stable lipid profile and neuronal functions. We confirmed behavior changes in IGF-1 injected APP/PS2 mice and analyzed hippocampus and adipose tissues for transcriptomic analysis. Our data suggested the alters of synapse- and neuroinflammation-related function in the hippocampus and the change of fat browning and cell

death signaling in adipose tissue of APP/PS2 mouse by IGF-1 treatment. We also obtained the list of long non-coding RNAs (lncRNAs) and circular RNAs (circRNAs) differentially expressed after the IGF-1 treatment and predicted their possible roles. Taken together, our study indicates the transcriptomic information both in hippocampus and adipose tissues in AD mouse model and suggests the function of IGF-1 in hippocampus and adipose tissues.

Disclosures: D. Jo: None. **J. Song:** None.

Poster

PSTR131. Altered Energy Homeostasis in Neurodegenerative Disorders

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR131.06/E18

Topic: C.02. Alzheimer's Disease and Other Dementias

Support: K99AG065645
R00AG065645

Title: Role of mitochondrial microRNAs in alzheimer's disease

Authors: *M. TORRES[1], B. SHARMA[2], S. KUMAR[2];
[1]Mol. and Translational medicine, TTUHSC El Paso, El Paso, TX; [2]Mol. and Translational medicine, TTUHSC EL Paso, EL Paso, TX

Abstract: Mitochondria plays crucial roles at synapses in providing synaptic energy and to maintain healthy synaptic and cognitive functions. Amyloid-beta and phosphorylated tau protein oligomers cause severe mitochondrial defects in Alzheimer’s disease (AD), that leads to the lack of synaptic energy and impaired synapse functions in AD. MicroRNAs (miRNAs) present within the mitochondria are involved in multiple mitochondrial activities and mitochondrial functions. Mitochondrial dysfunction is well established in AD; but the roles of mitochondrial miRNAs has not been determined in AD. Current study is focused on the identification of mitochondrial miRNAs in AD and to unveil their roles in disease pathogenesis. Mitochondria and cytosolic fraction were extracted from postmortem AD brains (n=5) and cognitively normal postmortem brains (n=5). Mitochondria purity was characterized by transmission electron microscopy and immunoblot analysis of mitochondrial markers. Further, total RNA was extracted from mitochondria and cytosolic fraction and subjected to miRNAs HiSeq analysis. MiRNAs high throughput analysis showed the deregulation of mitochondrial miRNAs in AD and control samples. We also found the miRNAs localization and differential expression in mitochondrial fraction relative to cytosolic fraction. Further, *in silico* bioinformatic analysis revealed the critical roles of mitochondrial miRNAs in several mitochondrial function and synaptic pathways in AD. Therefore, our study discovered some novel mitochondrial miRNAs, those could be the potential therapeutic target to retrieve mitochondrial and synaptic dysfunction in AD.

Disclosures: M. Torres: None. **B. Sharma:** None. **S. Kumar:** None.

Poster

PSTR131. Altered Energy Homeostasis in Neurodegenerative Disorders

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR131.07/E19

Topic: C.02. Alzheimer's Disease and Other Dementias

Support: NIA Grant AG050490
NIA Grant AG062548
NIA Grant AG054073
NIH Grant AG035982
Margaret “Peg” McLaughlin and Lydia A. Walker Opportunity Fund

Title: Investigating the relationship between mitochondrial DNA and Alzheimer's Disease biomarkers

Authors: *R. E. KEMNA[1], I. WEIDLING[5], C. LYSAKER[2], T. STROPE[3], V. CSIKOS DRUMMOND[6], P. KUECK[1], C. JOHNSON[4], C. S. JOHN[1], R. H. SWERDLOW[7], H. M. WILKINS[2], J. K. MORRIS[8];
[2]Univ. of Kansas Med. Ctr., [3]KUMC, [1]Univ. of Kansas Med. Ctr., Kansas City, KS; [4]Univ. of Kansas Med. Ctr., Lenexa, KS; [5]AbbVie, Cambridge, MA; [6]Kansas University, Lawrence, The Univ. of Kansas, Lawerence, KS; [7]Univ. Kansas Sch. Med., Leawood, KS; [8]KU Med. Ctr., KU Med. Ctr., Kansas City, KS

Abstract: Deficits in metabolism and mitochondrial function are contributing risk factors for the development of Alzheimer’s Disease (AD). These factors can affect the body’s ability to maintain proper cellular functions like proteostasis and mitophagy, among others. Mitochondria are unique from other organelles as they have their own genome (mtDNA), separate from nuclear DNA, which encodes for protein complexes involved in oxidative phosphorylation and cellular metabolism. To investigate how variations in mtDNA influence diseases such as AD, we used cytoplasmic hybrid (cybrid) cell models to investigate the relationship between mtDNA and proteins associated with AD pathology. These cybrid cell lines were generated from clinical research volunteers (n=11 AD, n= 8 cognitively healthy) enrolled into the Relationship of Energetics and Cognitive Trajectory study (KUMC IRB #03492). We analyzed cell lines for expression of AD-related proteins, cellular metabolic flux, and secretion of proteins. Hyperphosphorylation of tau protein is seen in AD, leading to pathogenic intracellular neurofibrillary tau-tangles; we observed elevated expression of pTau181 ($p=0.015$) in cybrids generated from subjects with AD when compared to cybrids from individuals without cognitive impairment. This suggests that mtDNA is related to cell functions that influence AD neuropathology. We also observed diagnostic differences in the protein most commonly associated with AD, amyloid-beta. Protein isolations from cell media showed differentially secreted amounts of amyloid-beta when comparing subjects with and without AD ($p<0.001$). These data, along with quantification of other proteins and analysis of cellular metabolic flux, suggest that mtDNA likely influences mitochondrial function and sequential disease-related

protein expression. Experiments are ongoing in a larger cohort of cybrid cells for further elucidation of the role of mitochondrial genetics in AD-related biomarkers.

Disclosures: **R.E. Kemna:** None. **I. Weidling:** None. **C. Lysaker:** None. **T. Strope:** None. **V. Csikos Drummond:** None. **P. Kueck:** None. **C. Johnson:** None. **C.S. John:** None. **R.H. Swerdlow:** None. **H.M. Wilkins:** None. **J.K. Morris:** None.

Poster

PSTR131. Altered Energy Homeostasis in Neurodegenerative Disorders

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR131.08/E20

Topic: C.02. Alzheimer's Disease and Other Dementias

Support: R01AG067330

Title: AdipoRon rescues intrinsic excitability and metabolic deficits caused by neurofibrillary tangle aggregation in neurons

Authors: *D. J. LASKY[1], E. R. MCGREGOR[2], O. J. RIPPENTROP[1], S. L. G. WRIGHT[1], R. M. ANDERSON[2], M. V. JONES[1];
[1]Neurosci., [2]Med., Univ. of Wisconsin, Madison, Madison, WI

Abstract: Alzheimer’s disease, with the hallmark accumulation of neurofibrillary tangles, induces neuronal and mitochondrial dysfunction. In recent years, AdipoRon, an adiponectin receptor agonist that activates AMPK signaling, has been found to alleviate both of these dysfunctions. We investigated how AdipoRon changed the intrinsic excitability and metabolic properties of neurons in mouse models of Alzheimer’s disease with the goal of connecting the mechanisms underlying these changes. Primary cortical neurons were isolated from the brains of postnatal day 0 Tau P301S (Tau) and APP/PS1 (Amy) mice. Tau transgenic (Tg) neurons were seeded with 3 µg/mL of pre-formed fibrils on day 7 *in vitro* to induce neurofibrillary tangle accumulation. Patch-clamp experiments were performed between 28-38 days *in vitro*. For AdipoRon experiments, cultures were incubated with 10 µM for 24 hours prior to patching. All recordings were done using HEPES-based extracellular solution and K-gluconate intracellular solution. Input resistance was assessed in voltage-clamp (−60 mV) and resting potential and spike firing were assessed in current-clamp, using D-APV (25 µM), DNQX (10 µM), and bicuculline methiodide (10 µM) to block synaptic transmission. Tau Tg neurons, compared to wild-type (WT), had significantly greater resistance (~88%) and a more depolarized resting potential (~23%). Tau Tg neurons spiked significantly less (~61%) in response to current applications than WT and were more likely to fire only a single spike (~86%). These differences were not found between Amy WT and Tg. When AdipoRon was applied to Tau Tg, both resistance and resting potential were rescued to levels similar to WT. AdipoRon application to Tau WT neurons did not significantly change their resistance or resting potential. Following the AdipoRon application, both Tau WT and Tg fired more spikes and were less likely to fire only a

single spike. These findings are consistent with Tau Tg having deficits in both resting K^+ and voltage-gated Na^+ channel function, that are rescued by AdipoRon. In separate experiments, we found that Tau Tg neurons experienced clearance of hyperphosphorylated tau following AdipoRon treatment. Additionally, Tau Tg neurons had reduced mitochondrial area and density compared to WT, and these impairments were similarly rescued by AdipoRon. In summary, Tau Tg neurons accumulated neurofibrillary tangles that correlated with deficits in both neuronal excitability and mitochondrial function. AdipoRon effectively cleared the hyperphosphorylated tau and restored, in some cases surpassing, wild-type excitability and mitochondrial properties.

Disclosures: D.J. Lasky: None. **E.R. McGregor:** None. **O.J. Rippentrop:** None. **S.L.G. Wright:** None. **R.M. Anderson:** None. **M.V. Jones:** None.

Poster

PSTR131. Altered Energy Homeostasis in Neurodegenerative Disorders

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR131.09/E21

Topic: C.02. Alzheimer's Disease and Other Dementias

Support: NIH grant R01AG071782
NIH grant R01AG084473

Title: Alzheimer's disease, metabolic syndrome, and aging have distinct effects on cerebral glucose transporters

Authors: *S. W. BARGER, Y. OU, J. SUNG;
Geriatrics, Univ. of Arkansas for Med. Sci., Little Rock, AR

Abstract: Attempts have been made to connect the neuropathological mechanisms of Alzheimer’s disease (AD) to those occurring in type-2 diabetes (T2D). However, the latter has been more definitively connected to increased risk of vascular dementia (VaD) than to AD, and persons with 2TD do not accrue higher indices of AD-related neuropathology (i.e., plaques and tangles). Nevertheless, impairment in cerebral metabolism of glucose (CMRglc) is a universal finding in AD and often occurs as a consequence of T2D. We previously discovered a change in glucose transporter 1 occurring specifically in astrocytes (GLUT1-A) in AD. Namely, cerebral GLUT1-A exhibits lower fractions of plasma membrane localization in humans with AD and in transgenic mice which accumulate amyloid β-peptide (Aβ). By contrast, most studies indicate that the hyperglycemia associated with T2D reduces overall expression of the GLUT1 contained in cerebrovascular endothelial cells (GLUT1-B). Here, we analyzed GLUT1-A, GLUT1-B, GLUT3, and GLUT4 in cerebrum of mice across a broad spectrum of mouse ages. We also analyzed the transporters in a mouse model of T2D (diet-induced obesity; DIO) and in the brains of humans who died with T2D. Total GLUT1-B expression was lower in humans diagnosed with T2D, regardless of their AD status. This was similar to mice with diet-induced obesity (DIO). Neither total levels nor membrane fractions of GLUT1-A were different in T2D or DIO.

Regarding aging, GLUT4 total levels were lower in 24-month-old mice compared to younger ages, but no changes were seen in GLUT1-A, GLUT1-B, or GLUT3. Membrane fractions of GLUT1-A and GLUT4 dropped between 7 and 12 months of age. Thus, AD, T2D, and normal aging of the brain produce distinct effects on the expression and subcellular trafficking of major glucose transporters. Metabolic syndrome and/or T2D suppress cerebral glucose transport from "outside-in"; i.e., they lower expression of GLUT1 in the cerebrovascular endothelium, perhaps as a consequence of exposure to elevated glucose. AD instead creates an "inside-out" suppression of glucose transport by impairing GLUT1-A plasma-membrane translocation. The diminished membrane translocation of GLUT4, observed in aging but not in AD, suggests the latter is not merely accelerated aging of the cerebrum. Because GLUT4 is insulin responsive, it seems that aging is more likely than AD to involve insulin resistance. Together, our findings suggest that the mechanisms producing CMRglc deficits are distinct in AD, T2D, and normal aging.

Disclosures: S.W. Barger: None. **Y. Ou:** None. **J. Sung:** None.

Poster

PSTR131. Altered Energy Homeostasis in Neurodegenerative Disorders

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR131.10/E22

Topic: C.02. Alzheimer's Disease and Other Dementias

Support: NIA Grant R01AG071782

Title: Estrogen replacement therapy restores cerebral glucose uptake and attenuates cognitive decline in ovariectomized Bri-Aβ42 mice

Authors: *N. D. SCHARTZ, Y. OU, J. SUNG, S. W. BARGER;
Geriatrics, Univ. of Arkansas for Med. Sci., Little Rock, AR

Abstract: Two thirds of individuals with late onset Alzheimer's disease (AD) are women, with advanced age and menopause being strong risk factors. Chronic decline in cerebral metabolic rate of glucose (CMRglc) precedes and, in individuals with mild cognitive impairment, predicts onset of AD cognitive decline and brain atrophy. Postmenopausal estrogen depletion may also contribute to deficiencies in CMRglc in women, thus increasing susceptibility to AD-associated cognitive decline. In the Bri-Aβ42 (Aβ-Tg) mouse model, we previously reported that young female mice are protected against metabolic and cognitive decline compared to males. Thus, we hypothesize that estrogens in female Aβ-Tg mice protect against amyloid-induced pathology. WT and Aβ-Tg female mice underwent bilateral ovariectomy (OVX) or sham surgery at approximately 6 weeks of age. After 2 weeks, mice were implanted with a timed-release subcutaneous pellet containing 17β-estradiol (βE2) or a placebo pellet (n=10-13/group). Two weeks after implantation, a subset of mice (n=8) were assessed for long-term spatial memory testing in an IntelliCage (IC). Mice were trained for one week to alternate between two opposite

corners of the cage to access drinking water. At 16 weeks of age, a subset of mice underwent glucose tolerance tests (GTT) and a 2-deoxyglucose (2DG) assay of CMRglc. Sham WT and Aβ-Tg mice significantly increased the correct pattern rate over 7 days of training in IC (Two-way ANOVA, $p < 0.05$). OVX Aβ-Tg mice failed to improve over time, but OVX Aβ-Tg with βE2 had significantly greater correct responses as early as Day 2 of training. Analysis of the difference in performance on Day 1 vs. Day 2 showed that WT, Aβ-Tg, and Aβ-Tg-OVX-E2β mice all significantly improved by Day 2, whereas Aβ-Tg-OVX only changed by 0.72%. GTT results varied by treatment and not genotype; OVX mice of both genotypes were most impaired, while βE2 supplementation protected both genotypes. Finally, mice were injected intravenously with [^{3}H]2DG to measure cerebral glucose uptake. Aβ-Tg OVX mice had significantly lower levels of 2DG uptake compared to their WT counterparts (Two-way ANOVA, $p < 0.01$), while OVX WT mice had similar levels as sham-operated WT, indicating interacting effects of estrogen depletion and amyloid accumulation on CMRglc. Loss of estrogen production during menopause has been shown to precede cognitive decline in AD, while treatment with HRT may attenuate the onset of these symptoms. Here, we show that loss of estrogen in a model predisposed to amyloidosis may affect cognitive function via glucose metabolism.

Disclosures: **N.D. Schartz:** None. **Y. Ou:** None. **J. Sung:** None. **S.W. Barger:** None.

Poster

PSTR131. Altered Energy Homeostasis in Neurodegenerative Disorders

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR131.11/E23

Topic:

Support: R01AG068330

Title: Neuronal ATP sensitive potassium (KATP) channel activity couple's peripheral and brain metabolism to effect sleep.

Authors: *N. CONSTANTINO[1], C. M. CARROLL[4], R. IRMEN[2], N. A. L'ITALIEN[2], D. SATISH[2], J. A. SNIPES[2], L. A. JOHNSON[3], S. L. MACAULEY[2];
[2]Dept. of Physiol., [3]Dept. of Physiology, Sanders Brown Ctr. on Aging, [1]Univ. of Kentucky, Lexington, KY; [4]Wake Forest Sch. of Med., Winston Salem, NC

Abstract: ATP sensitive potassium (KATP) channels act as metabolic sensors to regulate cellular excitability. We demonstrated that neuronal KATP channels are composed of Kir6.2 subunits, and are highly expressed on excitatory and inhibitory neurons (Grizzanti et al, 2022). Mechanistically, we demonstrated that KATP channels couple changes in peripheral and cerebral metabolism with neuronal activity. Here, we extend these studies to explore how KATP channels contribute to excitatory/inhibitory (E/I) balance in the brain to impact sleep. How KATP channels coordinate sleep, metabolism, and neuronal activity was examined using the Kir6.2-/- mice, a mouse model lacking neuronal KATP channel activity. Intracranial biosensors measuring

brain interstitial fluid (ISF) glucose and ISF lactate concurrent with EEG/EMGs were implanted into the hippocampus of wild type (WT) and Kir6.2-/- mice. Diurnal rhythms of ISF glucose, ISF lactate, and EEG/EMG were recorded for 72 hours. Mice were injected with saline, glucose, or glibenclamide to determine effects of metabolic challenges on ISF glucose, ISF lactate, and sleep/wake cycles. Quantitative EEG analysis and sleep staging was performed and correlated with ISF glucose and lactate over the 24 hour light/dark period. WT and Kir6.2 -/- mice were also oral gavaged with stable isotope tracer 13C-glucose and sacked for brain tissue analysis of glucose utilization. We found that WT mice have diurnal fluctuations in ISF glucose and lactate, with increases during the dark cycle when mice are awake and decreases in the light cycle when mice are asleep. In Kir6.2-/- mice lacking neuronal KATP channel activity, diurnal fluctuations in ISF glucose and lactate are lost. Kir6.2-/- mice spent more time in NREM sleep and less time awake than WT mice. Furthermore, reductions in absolute EEG power suggest changes in the brain's E/I balance. Kir6.2-/- mice were unresponsive to metabolic challenges, demonstrating that ISF lactate, a metabolic trigger for wakefulness, is controlled by KATP channels. 13 C-glucose isotope tracer analysis showed that total abundance of neurotransmitters was similar in both WT and 6.2 -/- mice, but that there were differences in %fractional enrichment of labeled versus unlabeled glucose being shunted to different pathways.

Disclosures: **N. Constantino:** None. **C.M. Carroll:** None. **R. Irmen:** None. **N.A. L'Italien:** None. **D. Satish:** None. **J.A. Snipes:** None. **L.A. Johnson:** None. **S.L. Macauley:** None.

Poster

PSTR131. Altered Energy Homeostasis in Neurodegenerative Disorders

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR131.12/E24

Topic: C.02. Alzheimer's Disease and Other Dementias

Support: NIH Grant RO1AG048232
NIH Grant 1P50 AG047366 (KIA)
NIH Grant RF1AG058047
Paul & Daisy Soros Foundation
Gerald J. Lieberman Fellowship

Title: Restoring hippocampal glucose metabolism rescues cognition across Alzheimer's disease pathologies

Authors: *P. S. MINHAS[1], K. I. ANDREASSON[2];
[1]Neurol. & Neurolog. Sci., Stanford Univ., San Francisco, CA; [2]Neurol. & Neurolog. Sci., Stanford Univ., Stanford, CA

Abstract: Impaired cerebral metabolism is a pathologic feature of Alzheimer's disease (AD) and recent proteomic studies highlight a disruption of neuroglial carbohydrate metabolism with disease progression. Here, we report that inhibition of indoleamine-2,3-dioxygenase (IDO1),

which metabolizes tryptophan to kynurenine, rescues hippocampal memory function and plasticity in AD models of amyloid and tau pathology by restoring astrocytic metabolic support of neurons. Activation of IDO1 in astrocytes by amyloid-beta42 and tau oligomers, two major pathological effectors in AD, suppresses glycolysis in an AhR-dependent manner, and pharmacological IDO1 inhibition restores glycolysis and lactate production by mouse and human iPSC derived astrocytes. In vivo, metabolomic analysis and MALDI-MS demonstrate that IDO1 inhibition restores hippocampal glucose metabolism, spatial memory, and synaptic plasticity in both amyloid-producing APPSwe-PS1ΔE9 and 5X FAD mice as well as in tau-producing P19S mice. IDO1 blockade rescues hippocampal long-term potentiation (LTP) in a monocarboxylate transporter (MCT)-dependent manner, indicating that IDO1-mediated suppression of lactate production disrupts astrocytic metabolic support of neurons. Indeed, in vitro mass-labeling demonstrates that IDO1 regulates astrocyte generation of lactate that is then transported to neurons. In co-cultures of astrocytes and neurons derived from AD subjects, deficient astrocyte lactate transfer to neurons was normalized with IDO1 inhibition and improved neuronal glucose metabolism. Taken together, these findings suggest that IDO1-mediated disruption of astrocytic support of neurons occurs across both amyloid and tau pathologies, and that IDO1 inhibitors currently being evaluated for cancer treatment could be also considered for treatment of amyloid- and tau-mediated neurodegenerative diseases

Disclosures: P.S. Minhas: None. **K.I. Andreasson:** None.

Poster

PSTR132. Preclinical Strategies for Alzheimer's Disease

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR132.01/E25

Topic: C.02. Alzheimer's Disease and Other Dementias

Support: NIH T32GM139779
NIH R01DA052340

Title: Assessing an Isoform-Selective Heat Shock Protein 90-Beta Inhibitor for the Preventative Treatment of Alzheimer’s Disease in 5xFAD Mice

Authors: *B. D. K. GRATREAK[1], M. A. SERWETNYK[2], T. L. D'AMICO[2], B. S. J. BLAGG[2], J. M. STREICHER[3];
[1]Univ. of Arizona Grad. Interdisciplinary Program In Neurosci., Tucson, AZ; [2]Dept. of Chem. and Biochem., Univ. of Notre Dame, Notre Dame, IN; [3]Pharmacol., Univ. of Arizona, Tucson, AZ

Abstract: The incidence of Alzheimer’s Disease (AD) increases with age and results in catastrophic progressive neurodegeneration and memory loss. After decades of failed clinical trials, promising but controversial new immunotherapies have recently been introduced that moderately slow cognitive decline after diagnosis of mild-to-moderate dementia, but there is no

cure or preventative treatment for AD. Yet-unknown driving mechanisms that underly AD pathology and dementia and immune activation appear to be key. Here, we report the preventative rather than reversive arm of our studies to modify tightly evolutionarily conserved molecular chaperone protein networks that are closely tied to systemic immunological processes by selectively inhibiting an isoform of Heat Shock Protein 90 (Hsp90) in transgenic 5xFAD mice that model rapid AD progression. Non-selective Hsp90 inhibitors historically show efficacy in improving AD pathology with a strong link to anti-inflammatory immune modulation of microglia signaling but were halted in clinical use by toxic side effects including hepatotoxicity and retinal degeneration. Literature suggests toxicity of pan-Hsp90 inhibitors is likely driven by effects of Hsp90α isoform inhibition. Our lead compound NDNB-01 is >333 fold selective for Hsp90β, previously shown efficacy in other mouse models at 1 mg/kg by translatable routes of administration, penetrates the central nervous system, and is orally bioavailable. By selectively inhibiting Hsp90β with NDNB-1 and avoiding Hsp90α inhibition, we predict the ability to achieve the benefits of pan-Hsp90 inhibition with fewer side effects. We hypothesize that Hsp90β-selective inhibition will reduce AD pathology in the 5X-FAD mouse model by immune modulation, specifically by decreasing inflammatory microglial activation. Two-month-old female and male 5xFAD mice were treated daily by subcutaneous injection of NDNB-1 (1mg/kg) and tested through biweekly open field test, novel object recognition test, and overnight nestbuilding assays. Our behavior data thus far suggests that Hsp90β inhibition is conferring a significant cognitive benefit to long-term 7-day novel object memory retention in NDNB-1-treated 5xFAD mice compared to vehicle-treated mice at thirteen weeks of treatment ($p=.0291$) without readily apparent side effects, though an organ panel screen for toxicity is pending. We further report our measurements of AD pathology in mouse brain tissue via Aβ42 levels, BACE1 activity, and CD11b/GFAP, as well as stimulation with Aβ42 peptide of NDNB-1 treated BV-2 microglial cells *in vitro* and measurement of their microglial polarization (CD32/CD86 vs. CD163/CD206).

Disclosures: **B.D.K. Gratreak:** None. **M.A. Serwetnyk:** None. **T.L. D'Amico:** None. **B.S.J. Blagg:** None. **J.M. Streicher:** E. Ownership Interest (stock, stock options, royalty, receipt of intellectual property rights/patent holder, excluding diversified mutual funds); Teleport Pharmaceuticals, LLC, Botanical Results, LLC.

Poster

PSTR132. Preclinical Strategies for Alzheimer's Disease

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR132.02/E26

Topic: C.02. Alzheimer's Disease and Other Dementias

Support: Ressler Family Foundation
NIH R37NS102185
The Dr. Miriam and Sheldon G. Adelson Medical Research Foundation
UCLA Eli and Edythe Broad Center of Regenerative Medicine and Stem

Cell Research
Steffy Family Trust

Title: Intercellular Signaling Pathways as Therapeutic Targets for Vascular Dementia Repair

Authors: *M. TIAN[1], R. KAWAGUCHI[1], Y. SHEN[2,3], S. MAGAKI[1,4], H. V. VINTERS[1,4], J. D. HINMAN[1], L. DE BIASE[5], Y. ZHANG[6], A. J. SILVA[2,3], S. T. CARMICHAEL[1]; [1]Neurol., [2]Neurobiology, Psychiatry and Psychology, [3]Integrative Ctr. for Learning and Memory, [4]Pathology and Lab. Med., [5]Physiol., [6]Psychiatry and Biobehavioral Sci., The David Geffen Sch. of Med. at UCLA, Los Angeles, CA

Abstract: Vascular dementia (VaD) constitutes approximately 25% of the total dementia population and imposes a substantial burden on the aging brain. Due to the lack of direct treatment, it is important to identify mechanisms for repair and recovery. To achieve this goal, the current study employed a mouse VaD model that replicates human VaD in multiple aspects including circuit damage by cerebral white matter ischemia, as well as memory and motor deficits. By leveraging cell type-specific RNA-Seq analysis, a unique transcriptomic profile of the white matter in the neurovascular niche was identified. This was interfaced with a human snRNA-Seq dataset in human VaD. By employing a custom-made database encompassing 4053 human and 2032 mouse ligand-receptor interactions in VaD, altered intercellular signaling pathways shared between human and mouse were identified as the potential targets for VaD treatment, highlighting ECM and GPCRs as major candidates. Microglia emerged as one of the key players in these pathways. One of the potential targets, CD39-A3AR signaling between endothelial cells and microglia, is significantly impaired in both aged mouse VaD model and human VaD patients. Treatment with an A3AR-specific agonist, which is under a clinical phase III for psoriasis, resulted in significant tissue and behavioral improvements in the mouse VaD model, underscoring the potential of targeting CD39-A3AR as a viable therapeutic strategy for VaD. Collectively, this study presents cutting-edge approaches to investigate the mechanism of VaD treatment, and identifies novel targets in VaD that may open new avenues for therapies.

Disclosures: M. Tian: None. **R. Kawaguchi:** None. **Y. Shen:** None. **S. Magaki:** None. **H.V. Vinters:** None. **J.D. Hinman:** None. **L. De Biase:** None. **Y. Zhang:** None. **A.J. Silva:** None. **S.T. Carmichael:** None.

Poster

PSTR132. Preclinical Strategies for Alzheimer's Disease

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR132.03/E27

Topic: C.02. Alzheimer's Disease and Other Dementias

Support: HU CoM Bridge Grant 2021-2022

Title: Effect of high salt diet on memory and behavior in apolipoprotein E4 expressing mice

Authors: R. ABDULMONIEM[1], M. RIVERS[1], G. CARTER[1], D. LEE[2], S. J. KHUNDMIRI[4], ***J. N. O'NEIL**[3];
[1]Howard Univ., Washington, DC; [3]Dept. of Physiol. & Biophysics, [2]Howard University, Col. of Med., Washington, DC; [4]Physiol., Howard Univ. Col. of Med., Washington, DC

Abstract: Several reports have demonstrated that the expression of Apolipoprotein E4 (APOE4) allele in humans increases the genetic risk of Alzheimer disease (AD). AD is characterized by a progressive decline in cognitive functions and memory. However, there is complete lack of the association of AD with dietary habits and any other comorbidities especially hypertension. Therefore, we hypothesized that high dietary salt intake will result in impaired spatial learning and impairment in mice expressing human APOE4 allele. To address this hypothesis, we used mice expressing human APOE4 or control APOE3 alleles exclusively in brain.Young adult male and female mice age 5–7month-old (n=6 in each group) were fed a 4% NaCl (high salt) or a 0.1% NaCl (normal salt) diet for 4 weeks. Learning and memory indices were determined using the Barnes maze test. APOE4 mice had decreased primary latency and primary errors when fed a low salt diet when compared to APOE3 mice. However, APOE4 mice showed increased primary latency and primary errors when fed a high salt diet. Our data suggest that low salt has beneficial effects on spatial learning and memory while high salt increases spatial deficits in learning and memory in APOE4 mice.

Disclosures: R. Abdulmoniem: None. **M. Rivers:** None. **G. Carter:** None. **D. Lee:** None. **S.J. Khundmiri:** None. **J.N. O'Neil:** None.

Poster

PSTR132. Preclinical Strategies for Alzheimer's Disease

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR132.04/E28

Topic: C.02. Alzheimer's Disease and Other Dementias

Support: NIH AG067473
VA RX003865 SPiRE Grant
NIH NS114221

Title: Preventive and preconditioning memantine treatment for the comorbidity of stroke and Alzheimer’s disease

Authors: H. GU[1], M. Q. JIANG[1], S. POURKHODADAD[2], L. WEI[3], ***S. YU**[1];
[1]Emory Univ., Atlanta, GA; [2]Emory Univ., ATLANTA, GA; [3]Dept. of Anesthesiol., Emory Univ., Atlanta, GA

Abstract: Alzheimer’s disease (AD) represents most dementia cases; ischemic stroke is a leading cause of death and disability in the same population. Although AD and stroke are different diseases, they are significant risk factors for each other. AD patients have high risks (>50%) for stroke; while one-third of stroke patients develop post-stroke dementia (PSD) and

AD-like pathology such as β-amyloid deposition. The comorbidity of these two diseases in the same patients calls for specific research attention and represents an eminent medical gap. AD and stroke share common pathophysiological mechanisms such as NMDA receptor (NMDAR) hyperactivity, excitotoxicity, detrimental inflammation, and neurovascular destruction. We recently identified that deficiency of the NMDAR GluN3A subunit is a pathogenic factor in sporadic AD development. The present investigation is to test the hypothesis that early treatment of the NMDA receptor antagonist memantine (MEM) is a disease-modifying therapy for preventing AD development and, in the same individuals, a preconditioning treatment antagonizing ischemic attack that is assured to attack at least 50% of AD cases. MEM selectively inhibits extrasynaptic NMDARs. Considering the NMDAR-mediated excitotoxicity and degenerative cascades in both diseases, we propose that early MEM treatment can provide two main benefits: 1) to mitigate chronic excitotoxicity underlying AD development, and 2) to prime (precondition) the AD brain for enhanced tolerance against acute excitotoxicity of ischemic stroke than may occur at any given time. In the GluN3A knockout mouse and the AD transgenic 5XFAD mouse, oral administration of MEM (10 mg/kg/day in drinking water) for 3 months during preclinical stage of AD resulted in significant benefits of attenuating the infarct formation induced by focal ischemic stroke. AD mice received the MEM treatment before and after stroke performed significantly better in multiple tests for sensorimotor, psychological and cognitive functions. The MEM treatment suppressed inflammatory factors and cells. The study shows the dual benefits of the clinical drug MEM in two AD mouse models before and after ischemic stroke.

Disclosures: H. Gu: None. **M.Q. Jiang:** None. **S. Pourkhodadad:** None. **L. Wei:** None. **S. Yu:** None.

Poster

PSTR132. Preclinical Strategies for Alzheimer's Disease

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR132.05/E29

Topic: C.02. Alzheimer's Disease and Other Dementias

Support: RF1 AG 059321

Title: The effect of plasma exchange in Amyloid beta (Aß) pathology

Authors: *S. KOERICH, S. RAMIREZ, N. ASTUDILLO CORRAL, C. SOTO;
The Univ. of Texas Hlth. Sci. Ctr. at Houston (UTHealth), Houston, TX

Abstract: Introduction: Alzheimer's disease (AD) is the most common type of dementia, characterized by neuronal death, synaptic alterations, neuroinflammation, and cerebral protein aggregates. Evidence suggests that peripheral Aβ plays a crucial role in AD pathogenesis. Previous studies have reported an equilibrium between cerebral and peripheral pools of Aβ. Additionally, impairment of peripheral Aβ clearance is believed to be linked to accelerated brain

amyloidosis and increased risk of developing dementia. Despite decades of effort to understand AD genetic and molecular causes, there is not yet an effective treatment or cure that delays the disease onset or slows the progression. Thus, the discovery of novel therapeutic strategies is a high priority. Our previous study showed that serial whole blood exchange in AD mice model led to prophylactic and therapeutic benefits in terms of reducing amyloid pathology, brain inflammation and improvement of memory. **Aim:** In the present study, we investigated the possibility of using repetitive plasma exchange, an FDA-approved procedure, as a way to clear the circulation from toxic species, including Aβ oligomers. Our overarching hypothesis is that AD can be treated at the periphery by removing Aβ from plasma. **Methods:** 300 μL of blood was obtained once a month from APP/PS1 mice from the jugular vein. Subsequently, the blood was centrifuged to remove the plasma, and the same volume was replaced with 5% albumin in saline and reinfused into the jugular vein. The procedure was performed 4 times between 3 to 6 months, and animals were humanely euthanized at 7 months. For biochemical and histological analysis, brains were dissected after transcardiac brain perfusion. The right hemisphere was fixed for histological examinations, and the left hemisphere was snap-frozen in liquid nitrogen. For histology, 10 μm sections were stained with thioflavin S (ThS), or incubated with anti-Aβ antibody 4G8 to analyze amyloid burden and number of Aβ plaques. All images were analyzed using NIH Image J software. Plasma concentrations of Aβ was measured by ELISA. **Results:** Our results show that monthly plasma exchange procedure effectively maintained Aβ in plasma at basal levels and reduced the amyloid burden and the number of amyloid deposits in 7-month-old mice. **Conclusions:** Currently, the precise mechanism through which plasma exchange reduces amyloid pathology in this model remains unclear. The decrease in cerebral amyloid deposition occurred in parallel with a reduction in the concentration of Aβ in plasma, indicating that the mobilization of Aβ from the brain to the bloodstream may play a role reducing the amyloid burden induced by plasma exchange.

Disclosures: **S. Koerich:** None. **S. Ramirez:** None. **N. Astudillo Corral:** None. **C. Soto:** None.

Poster

PSTR132. Preclinical Strategies for Alzheimer's Disease

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR132.06/Web Only

Topic: C.02. Alzheimer's Disease and Other Dementias

Title: Cannabidiol-based nanoemulsions as a potential treatment for Alzheimer's disease

Authors: *T. ZIENELDIEN, *J. KIM, Y. WANG, O. FIHURKA, N. SHEN, Q. ZHOU, C. CAO;
Univ. of South Florida, Tampa, FL

Abstract: Alzheimer’s disease (AD) is widely acknowledged as having a multifactorial pathogenesis, indicating that a combinational therapy approach is necessary to address the various factors involved and enhance the overall efficacy of treatment. Many studies have

elucidated the potential of melatonin and insulin in reversing and preventing AD-related cognitive decline, amyloid β (Aβ)-incited neuroinflammation, mitochondrial instability, and neuronal loss. Similarly, there has been increasing evidence of the potential of cannabidiol (CBD) to slow the progression of AD symptoms by reducing Aβ accumulation and tau hyperphosphorylation, positioning CBD as a compelling candidate for inclusion in AD therapeutic strategies. The objective of this research was to develop a nasal nanoformulation composed of melatonin, insulin, and CBD for AD therapy. In our investigation, we evaluated the ability of the MIC (melatonin 0.04 mg/kg; insulin 0.008 mg/kg; CBD 0.2 mg/kg) and MC (melatonin 0.04 mg/kg; CBD 0.2 mg/kg) anti-AD nanoformulations to protect aged APP/PS1 from memory decline and AD-related pathological changes. 12-month-old APP/PS1 mice underwent spatial memory testing on the radial arm water maze (RAWM). Tests were performed before and after the 3-month once-daily intranasal treatment with MIC or MC nanoemulsion (non-transgenic (NTG) group n=11, transgenic (TG) group n=6; MIC group n=6; MC group n=7). Daily intranasal administration of MIC and MC significantly enhanced spatial learning memory in comparison to transgenic control mice ($p<0.01$ and $p<0.001$, respectively). Biochemical analyses revealed a significant beneficial impact of MIC and MC treatment on the neuropathological alterations compared to the untreated TG control group; further, both nanoformulations can balance mitochondrial dynamics. Immunohistochemical (IHC) results show that the MC treatment has a beneficial impact on reversing the neuropathologic changes of AD, such as Aβ accumulation and microglial activation. At the same time, the MIC treatment elevated the number of mature neuronal cells. Overall, the MIC and MC nanoformulations are promising combination therapeutics for AD, displaying notable neurocognitive effects and positively impacting AD-associated pathologies, indicating they are potential candidates for long-term AD treatment.

Disclosures: **T. Zieneldien:** None. **J. Kim:** None. **Y. Wang:** None. **O. Fihurka:** None. **N. Shen:** None. **Q. Zhou:** None. **C. Cao:** E. Ownership Interest (stock, stock options, royalty, receipt of intellectual property rights/patent holder, excluding diversified mutual funds); University of South Florida.

Poster

PSTR132. Preclinical Strategies for Alzheimer's Disease

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR132.07/Web Only

Topic: C.02. Alzheimer's Disease and Other Dementias

Support: VIEP-BUAP 2023
CF-2023-G-597

Title: Cannabidiol (cbd) improves memory in lesioned rats with intracerebroventricular oAB$_{25-35}$ injection.

Authors: *E. MACUIL CHAPULI[1,3], E. MARTÍNEZ JUÁREZ[3], T. TZOMPANTZI JUÁREZ[3], I. D. LIMÓN[2,3], A. PATRICIO MARTÍNEZ[2,4];
[1]Neuropharm. laboratory, [2]Benemérita Univ. Autónoma de Puebla, Puebla, Mexico;
[3]Neuropharm. laboratory FCQ-BUAP, Puebla, Mexico; [4]Fac. of Biol. Sci., Puebla, Mexico

Abstract: Alzheimer's disease (AD) is the most common neurodegenerative condition, primarily among older adults, that jeopardizes the global population as life expectancy increases. The two major cellular markers in Alzheimer's are characterized by the presence of Aβ plaque depositions and neurofibrillary Tau tangles, coupled with behavioral changes and progress cognitive impairment, altogether account for the principal symptoms of the dementia caused by the disease. Currently, there are no existing treatments to prevent or to stop neurodegeneration and progression of dementia. Therefore, it is necessary to explore new treatments and pharmacological alternatives. Cannabidiol (CBD) comes up as a pharmacological option to cope with harmful symptoms due to its pleiotropic properties, having the capacity to interact with different endogenous signaling systems. The aim of this study was to evaluate the effect of CBD on memory performance in Morris water maze after $A\beta_{25-35}$ oligomer ($oA\beta_{25-35}$) injection. We used $oA\beta_{25-35}$ to cause an Alzheimer's animal model as oligomer produces most of the detrimental effects in brain cells. A single Aβ intracerebroventricular injection was applied to assess male Wistar rats administrated with CBD or vehicle throughout 24 days. Four treatments were experimentally tested, each consisted of rats receiving either $oA\beta_{25-35}$ + CBD n=8, $oA\beta_{25-35}$ + Veh n=9, $oA\beta_{35-25}$ + CBD n=9, or $oA\beta_{35-25}$ + Veh n=10. CBD administration had an effect on animal's long-term memory, improved scape latency and increased number of crosses to the scape platform in Morris water maze, in contrast, rats administrated only with $oA\beta_{25-35}$ took longer to find the platform zone and had fewer crosses. In conclusion, individuals under CBD treatment showed improved behavioral performance suggesting a possible neuroprotective role of CBD thus having a potential therapeutic use in AD pathology.

Disclosures: E. Macuil Chapuli: None. **E. Martínez Juárez:** None. **T. Tzompantzi Juárez:** None. **I.D. Limón:** None. **A. Patricio Martínez:** None.

Poster

PSTR132. Preclinical Strategies for Alzheimer's Disease

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR132.08/E30

Topic: C.02. Alzheimer's Disease and Other Dementias

Support: R21AG073934 NIH/NIA
Helen Diller Family Comprehensive Cancer Center Survivorship and Symptom Science (SASS) Research Hub Award
Bakar Aging Research Institute Award
Berkelhammer Award for Excellence in Neuroscience
RF1 AG064170

Title: Neuropathologic analysis of ionizing radiation effects in mouse models of neurodegeneration

Authors: A. OYEWOLE[1], **M. LEE**[2], J. YANG[2], Y. SEI[2], J. SIMMS[2], *K. NAKAMURA[2], J. L. NAKAMURA[1];
[1]Radiation Oncology, UCSF, San Francisco, CA; [2]Neurolog. Dis., Gladstone Inst., San Francisco, CA

Abstract: Ionizing radiation (IR) exposure can occur through diagnostic and therapeutic medical procedures. A serious complication of brain exposure to IR is cognitive decline, although the exact mechanisms of brain injury are unclear. Furthermore, whether certain individuals are more susceptible to radiation brain injury is unknown. We aim to investigate the effect of radiation exposure on brains also at risk for neurodegeneration. In this study, we exposed an Alzheimer's mouse model (mutAPP knock-in on a human ApoE3 versus E4 KI background) to brain-only radiation (0 Gy, 3 Gy x 2 doses, 5 Gy x 2 doses) and analyzed brains collected at 1 month or 4 months post radiation. We then performed immunohistochemistry (IHC) on brain tissue slices to probe for key pathological hallmarks in Alzheimer's disease (AD) - associated proteins including extracellular amyloid beta plaques, phosphorylated tau, and ApoE. With confocal microscopy imaging, we quantified AD histopathologic markers in mice. At 1 month post-radiation, hippocampal amyloid beta plaques were reduced in the ApoE4/APP genotype at both radiation doses. Radiation had no impact on ApoE levels at four months, with additional analyses ongoing. Neurobehavioral testing at one and four months are currently being analyzed. These initial histopathologic findings suggest that radiation exposure to the brain influences AD pathology in specific genetic contexts and in a dose-dependent manner.

Disclosures: A. Oyewole: None. **M. Lee:** None. **J. Yang:** None. **Y. Sei:** None. **J. Simms:** None. **K. Nakamura:** None. **J.L. Nakamura:** None.

Poster

PSTR132. Preclinical Strategies for Alzheimer's Disease

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR132.09/E31

Topic: C.02. Alzheimer's Disease and Other Dementias

Support: AMED Brain/MINDS (JP19dm0207079h to MK)
JSPS Grant-in-Aid for Scientific Research (19H01152, 20H00575 and 23H00522 to M.K.)
JST SPRING (JPMJSP2128 to DJ)

Title: Cognitive inflexibility of Alzheimer's model: App^{NL-G-F} mice exhibit delayed adaptation to altered contingency in case of no prior knowledge

Authors: *D. JOHO[1], T. SUZUKI[1], E. N. MINAKAWA[3], T. FURUSE[4], T. SAITO[5], H. SASAGURI[6], T. C. SAIDO[7], M. KAKEYAMA[2];

[1]Lab. for Envrn. Brain Science, Grad. Sch. of Human Sci., [2]Lab. for Envrn. Brain Science, Fac. of Human Sci., Waseda Univ., Tokorozawa, Japan; [3]Dept. of Neurophysiol., Natl. Inst. of Neuroscience, Natl. Ctr. of Neurol. and Psychiatry, Kodaira, Japan; [4]Mouse Phenotype Analysis Div., RIKEN BioResource Res. Ctr., Tsukuba, Japan; [5]Dept. of Neurocognitive Sci., Inst. of Brain Science, Nagoya City Univ. Grad. Sch. of Med. Sci., Nagoya, Japan; [6]Dementia Pathophysiology Collaboration Unit, [7]Lab. for Proteolytic Neurosci., RIKEN Ctr. for Brain Sci., Wako, Japan

Abstract: Alzheimer's disease (AD) is the most common neurodegenerative disease. More than 100 mice models have been generated, and in many cases, hippocampal-dependent memory task such as memories of a specific location have been investigated. However, the hippocampal atrophy in patients with AD and such severe memory impairment, that is, memory loss of their own bedroom, are prominently exhibited as progressive symptoms. A behavioral task paradigm is required that allows quantitative detection of not only simple learning and memory impairment but also higher-order functions, such as cognitive flexibility, which is often found in patients even with mild cognitive impairment. We previously established a cognitive flexibility task using IntelliCage, a fully automated behavioral apparatus for group-housed mice. This task requires mice to acquire a spatial learning-based behavioral sequence as a habit and then to adapt reversal task (changes in locations of rewards) while maintaining the behavioral sequencing habits. In this cognitive flexibility task, mice show progressive improvement to altered contingency as the reversal changes were repeated, that is, mice form the reversal learning-set. In this study, the cognitive flexibility task was conducted in the 2nd generation APP knock-in mouse models to assess the adaptation speed to altered contingency (App^{NL-G-F}, n = 12 and App^{NL}, n = 10). App^{NL} mice, which harbor only the Swedish mutation, were used as controls. We divided the mice into two groups: in the first group, the flexibility task was conducted once at middle age (14 months); in the second group, the flexibility task was conducted twice (at 4 months and 14 months of age). The results showed that middle-aged App^{NL-G-F} mice required greater access to altered contingency than control mice as repeating reversal changes, whereas spatial learning ability was consistent with the controls. Intriguingly, cognitive flexibility was retained in the middle-aged App^{NL-G-F} mice who had completed the task previously (4 months old). In other words, cognitive inflexibility was observed in App^{NL-G-F} mice without prior knowledge but not in the mice with prior knowledge, indicating that the function of acquiring knowledge may decline prior to spatial learning and retention of acquired knowledge. Taken together, these results indicate that cognitive flexibility can be a good index for assessing early or preclinical AD stages.

Disclosures: D. Joho: None. **T. Suzuki:** None. **E.N. Minakawa:** None. **T. Furuse:** None. **T. Saito:** None. **H. Sasaguri:** None. **T.C. Saido:** None. **M. Kakeyama:** None.

Poster

PSTR132. Preclinical Strategies for Alzheimer's Disease

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR132.10/Web Only

Topic: C.02. Alzheimer's Disease and Other Dementias

Title: Therapeutic dendritic cell Vaccine Targeting Oligomeric amyloid-β in mouse model of Alzheimer’s disease

Authors: *Y. WANG, N. SHEN, H. YANG, X. LIN, C. CAO;
Univ. of South Florida, Tampa, FL

Abstract: Background: Immunotherapy targeting the oligomeric forms of amyloid-β (Aβ) may halt the progression of AD.Objective: Develop a peptide-sensitized dendritic cell (DCs) vaccine that can specifically target oligomeric Aβ and prevent autoimmune responses.Methods: The study utilized Alzheimer's disease mouse models and non-transgenic mouse models. Antibody expression assays, cytokine expression profile detection, characterization of antisera, and spatial memory behavioral tests before and after vaccination were conducted.Results: Epitope prediction indicated that Aβ mutation(PDM) could generate new T cell epitopes, enhancing the immune response in elderly patients and suppressing certain T cell epitopes of wild-type Aβ. The suppression of these T cell epitopes can help prevent the autoimmune response observed in some patients receiving the AN-1792 vaccine. PDM not only facilitated the sensitization of bone marrow-derived DCs to produce specific antibodies against oligomeric Aβ but also delayed memory impairment in the APP/PS1 mouse model. Importantly, the PDM peptide did not alter the natural immunomodulatory properties of DCs.Conclusion: The PDM vaccine may be safer for patients with impaired immune systems. Given the mounting evidence suggesting that the oligomeric form of Aβ is detrimental to neurons, the specificity of PDM antibodies for these "oligomeric" Aβ species may offer clinical benefits to patients with AD.

Disclosures: **Y. wang:** None. **N. Shen:** None. **H. Yang:** None. **X. lin:** None. **C. Cao:** None.

Poster

PSTR132. Preclinical Strategies for Alzheimer's Disease

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR132.11/E32

Topic: C.02. Alzheimer's Disease and Other Dementias

Support: EMBO ALTF 67-2022
Innosuisse – Swis Innovation Agency - Swiss Confederation-56184.1 IP-LS
EMBO ALTF 111-2021

Title: Enhancing mitophagy improves cognition in models of Alzheimer's disease

Authors: *S. RODRÍGUEZ LÓPEZ[1], Q. WANG[1], A. LALOU[1], T. I LIMA[1], K. DUGI[2], P. A. ANDREUX[2], J. AUWERX[1];
[1]Ecole Polytechnique Federale de Lausanne, Lausanne, Switzerland; [2]Vandria SA, Lausanne, Switzerland

Abstract: Mitophagy is the selective elimination of damaged mitochondria through autophagy, failure in which leads to the accumulation of dysfunctional mitochondria. Impaired mitochondrial network exacerbates neurodegenerative disorders such as Alzheimer's disease (AD) through multiple vicious cycles, including aggravating oxidative stress and fueling neuroinflammation. Strategies aiming to restore mitophagy functions can be used as a therapeutic intervention for AD. We have developed a new mitophagy booster drug with high brain penetration, referred to as VNA-318 hereafter. We investigated the therapeutic potential of VNA-318 administration in mouse models of AD. Our findings reveal that chronic administration of VNA-318 provides neuroprotective effects against amyloid-induced neuronal loss and neuroinflammation and rescues cognitive deficits. In addition, VNA-318 can improve short-term spatial memory in aged mice and young mice injected with LPS. We also assessed the safety profile of chronic administration of VNA-318 as a treatment for AD in preclinical models. These data demonstrate the efficacy of VNA-318 preclinically and position it as a new Lead candidate for both disease-modification and enhancement of cognition.

Disclosures: S. Rodríguez López: None. **Q. Wang:** None. **A. Lalou:** None. **T. I Lima:** None. **K. Dugi:** A. Employment/Salary (full or part-time); Vandria SA, EPFL Innovation Park, Building E, CH-1015 Lausanne, Switzerland. **P.A. Andreux:** A. Employment/Salary (full or part-time); Vandria SA, EPFL Innovation Park, Building E, CH-1015 Lausanne, Switzerland. **J. Auwerx:** B. Contracted Research/Research Grant (principal investigator for a drug study, collaborator or consultant and pending and current grants). If you are a PI for a drug study, report that research relationship even if those funds come to an institution.; Supported by Innosuisse grant 56184.1 IP-LS.

Poster

PSTR132. Preclinical Strategies for Alzheimer's Disease

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR132.12/E33

Topic: C.02. Alzheimer's Disease and Other Dementias

Support: NIH Grant R01-AG062469
Research Endowment Funds, Center for Drug Design

Title: Orally bioavailable ψ-GSH prodrugs as multimodal therapeutics for Alzheimer's disease

Authors: *S. S. MORE, S. RAO, W. XIE, J. MEINTS, S. VERMILYEA, M. K. LEE; Univ. of Minnesota, Twin Cities, Minneapolis, MN

Abstract: Advanced glycation end products (AGEs), resulting from oxidation of sugar molecules, induce oxidative stress and inflammation by covalently modifying a broad spectrum of biomolecules. The role of AGEs in the development and progression of aging-associated conditions, Alzheimer's disease (AD), is well established. In AD and in transgenic mouse models of AD, increase in AGE is, in part, results from reduced glutathione (GSH) and increased

oxidative stress. Reduced GSH also leads to reduced Glyoxylase1 (Glo1) function required to detoxify oxidized sugars/methylglyoxal (MG). To attenuate oxidative stress in AD and increase Glo1 activity in brain, we developed a metabolically stable glutathione analog, ψ-GSH, which is able to substitute for GSH in the Glo1 enzymatic reaction and retain its antioxidant potential. In APP/PS1 mice, intraperitoneal ψ-GSH effectively engaged Glo1 and halted disease progression in symptomatic animals. However, ψ-GSH has poor oral bioavailability because of the labile thiol group. We designed and synthesized prodrugs of ψ-GSH by masking the labile thiol and achieved desirable oral bioavailability. The prodrugs display superior plasma, liver and gastrointestinal stability when compared to the parent ψ-GSH. Evaluation of the lead prodrugs in an AD-like mouse model generated by intracerebroventricular (*i.c.v.*) injection of $A\beta^{1-42}$ peptide displayed their behavioral and biochemical benefits (Xie et al., J. Med. Chem, 2022, 65:14441). To test if prodrug **7** can attenuate AD pathology, "symptomatic" APP/PS1 mice at 10 months of age were orally administered the prodrug **7** for 2 months. Behavioral analysis at 12 months of age show that the prodrug **7** treatment improved spatial learning and memory. Biochemical analysis of brain tissues displayed marked reduction in oxidative stress and neuroinflammation. Glo1 pathway intermediates were concurrently found to be modulated, confirming the involvement of Glo1 in the neuroprotection offered by prodrug **7**. Quantitative neuropathology indicated significant reduction in Aβ plaque burden and reactive astrogliosis. The prodrugs countered progressive neurodegeneration as evidenced by their effects on cortical NAergic afferents, TH+ neuronal number and density. These studies showcase the utility of ψ-GSH-based agents for reversing or impeding the progression of AD-like pathology. In turn, further preclinical exploration of the glyoxalase pathway for the development of anti-AD therapy is clearly warranted.

Disclosures: S.S. More: None. **S. Rao:** None. **W. Xie:** None. **J. Meints:** None. **S. Vermilyea:** None. **M.K. Lee:** None.

Poster

PSTR132. Preclinical Strategies for Alzheimer's Disease

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR132.13/E34

Topic: C.02. Alzheimer's Disease and Other Dementias

Support: NIH AG054345
NIH AG065181

Title: Inpp5d enhances Ab uptake, mitigates behavioral outcomes by reducing Ab pathogenesis

Authors: P. B.-C. LIN[1], T. RICHARDSON[2], B. T. LAMB[4], ***A. L. OBLAK**[3];
[1]SNRI, IUSM, Indianapolis, IN; [3]Stark Neurosciences Res. Inst., [2]Indiana Univ. Sch. of Med., Indianapolis, IN; [4]Stark Neurosciences Res. Inst., Indianapolis, IN

Abstract: Alzheimer’s disease is a neurodegenerative disorder and the most common cause of dementia. Genetic studies implicate the involvement of microglia-mediated immune responses during disease progression. Importantly, inositol polyphosphate-5-phosphatase D (INPP5D) serves as a regulator of microglial functions and its variants have been identified as risk of late-onset AD. This study investigates the role of INPP5D in the pathogenesis of Alzheimer's disease. We found that increased levels of INPP5D are detected in the brains of AD patients, particularly in regions associated with amyloid plaque density. Similar findings are observed in a murine model of AD. We further explored the effects of INPP5D haplodeficiency in the 5xFAD mouse model, revealing that reduced INPP5D expression decreases amyloid plaque burden, improves behavioral deficits, and enhances microglial engagement to plaques. It also activates TREM2 signaling, suppresses proinflammatory cytokine release, and modulates functional pathways related to immune cell activation, cytokine production, protein degradation, memory, and synaptic plasticity. In vitro studies show that INPP5D inhibition increases fibrillar beta-amyloid uptake, reduces cytotoxicity, and alters functional pathways associated with phagocytosis, apoptosis, cytokine production, and the complement system. These findings suggest that INPP5D inhibition may protect against AD pathology and that targeting microglia-mediated immune responses through INPP5D antagonists could be a potential therapeutic approach for AD.

Disclosures: P.B. Lin: None. **T. Richardson:** None. **B.T. Lamb:** None. **A.L. Oblak:** None.

Poster

PSTR132. Preclinical Strategies for Alzheimer's Disease

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR132.14/E35

Topic: C.02. Alzheimer's Disease and Other Dementias

Support: NIH Grant R01AG068215

Title: A novel protocol for probing the effects of sleep and temperature in a mouse model of Alzheimer's disease

Authors: *J. WANG[1], D. IRADUKUNDA[2], A. BACHSTETTER[5], M. P. MURPHY[3], B. F. O’HARA[4], M. J. DUNCAN[5], S. SUNDERAM[2];
[2]Dept. of Biomed. Engin., [3]Sanders-Brown Ctr. on Aging, Dept. of Biochem., [4]Dept. of Biol., [1]Univ. of Kentucky, Lexington, KY; [5]Neurosci., Univ. of Kentucky Med. Sch., Lexington, KY

Abstract: Disordered sleep, which is common in Alzheimer’s disease (AD), may accelerate neuropathology, thus promoting a vicious cycle. In a previous study, we investigated the hypothesis that improving sleep through diurnal exposure to thermoneutral temperatures may slow disease progression in 3xTg-AD mice. We found that slow wave sleep (SWS) - the deepest stage of NREM sleep dominated by delta oscillations in the EEG - was significantly elevated ($p<0.05$) in the light phase for treated mice compared to controls even though the time spent in total sleep, NREM, REM, and wakefulness were unchanged. Furthermore, both Aβ40 and Aβ42

were significantly lower in the hippocampus, but not in the cortex, for the treated group. These findings imply that thermoneutral conditions might have some regional specificity in their effects, with implications for the cognitive and neuropathologic changes found in AD.To further test whether the effect of thermoneutrality on amyloid pathology is mediated by sleep changes rather than other non-specific physiologic effects of temperature, we conducted a new study with a modified design. APP/PS1 knock-in mice (6 m.o, male) were instrumented for EEG/EMG monitoring to score sleep. After a week-long baseline recording, they were divided into four groups: 1. SE (n=8), exposed to thermoneutral temperature (30°C) during the 12-hour light period; 2. SD (n=8), which received intermittent vibratory stimuli to disrupt sleep during the light period; 3. SExSD (n=9), which received sleep disruption along with thermoneutral exposure; and 4. CTRL (n=8), which received no treatment. The SExSD condition is meant to compensate for any effects of temperature that are not mediated by sleep enhancement in the SE condition through sleep disruption. All animal procedures were carried out with prior approval from the Institutional Animal Care and Use Committee (IACUC) of the University of Kentucky.After four weeks of treatment, the animals were euthanized, and the brains removed to assay amyloid-beta (Aβ) levels using ELISA. Wake, REM, NREM, and SWS within NREM were scored from the EEG/EMG to analyze sleep. In our preliminary analysis of the data, we find that SWS was increased up to four-fold in SE mice. However, a similar increase on average was observed in SExSD mice despite the attempted sleep disruption. These outcomes will be correlated with pathology when the biochemical assays become available.

Disclosures: J. Wang: None. **D. Iradukunda:** None. **A. Bachstetter:** None. **M.P. Murphy:** None. **B.F. O'Hara:** A. Employment/Salary (full or part-time); Signal Solution LLC. **M.J. Duncan:** None. **S. Sunderam:** None.

Poster

PSTR132. Preclinical Strategies for Alzheimer's Disease

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR132.15/E36

Topic: C.02. Alzheimer's Disease and Other Dementias

Title: Protective Effects of Apigenin on Neuronal Aging and Neurodegeneration in C. Elegans

Authors: *R. A. GRANT, L. H. PARK, D. WAHL, T. EMGE, T. J. LAROCCA;
Colorado State Univ., Fort collins, CO

Abstract: Aging is the primary risk factor for a variety of chronic diseases including Alzheimer's disease (AD). Although the underlying neuropathology of AD has been thoroughly investigated, no effective treatments have been found. One novel therapeutic agent that has gained interest for its potential to treat age-related disease is the naturally occurring flavonoid apigenin. The molecular mechanisms that underlie apigenin's therapeutic effects are reported to involve anti-inflammatory signaling, antioxidant function, cell cycle arrest, and apoptosis. To determine if apigenin is protective against aging and neurodegeneration (and to identify potential

underlying mechanisms), we studied the effects of apigenin in various *C. elegans* models. We found that apigenin extends healthy lifespan in pan-neuronal tau-expressing worms by ~10%, and that this lifespan extension is associated with enhanced neuromuscular function, as reflected by improved pharyngeal pumping in older apigenin-treated animals vs. untreated controls. We also found that apigenin protects against age-related neurodegeneration, as evidence by reduced blebbing and degeneration of key sensory (PVD) neurons in treated *C. elegans*. To extend on these initial findings, we are testing apigenin in wildtype and pan-neuronal amyloid beta (Aβ) expressing worms for its effects on lifespan and neuromuscular function. Also, to better understand the mechanisms underlying apigenin's therapeutic effects, we are preforming RNA-seq on treated animals and examining the compounds effects in transgenic *C. elegans* strains with fluorescent reporters reflecting oxidative stress, cell-cycle arrest, and inflammation-related signaling. Currently, our results suggest that apigenin may protect against age-related neurodegeneration and AD-associated protein accumulation in *C. elegans*, and our ongoing experiments will help elucidate the molecular mechanisms involved.

Disclosures: R.A. Grant: None. **L.H. Park:** None. **D. Wahl:** None. **T. Emge:** None. **T.J. LaRocca:** None.

Poster

PSTR132. Preclinical Strategies for Alzheimer's Disease

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR132.16/E37

Topic: C.02. Alzheimer's Disease and Other Dementias

Support: EMBO ALTF 111-2021
EMBO ALTF 67-2022
Human Frontier Science Program LT000731/2018-L

Title: Restoring mitochondrial homeostasis as a therapy for Alzheimer's disease

Authors: Q. WANG[1], S. RODRÍGUEZ-LOPEZ[1], T. Y. LI[1], J.-D. MOREL[1], M. NELSON[2], J. AUWERX[1];
[1]École Polytechnique Fédérale de Lausanne, Lausanne, Switzerland; [2]Applied Mol. Design LLC, Midway, UT

Abstract: Mitochondrial dysfunction is a conserved feature of normal aging and many age-related chronic diseases including neurodegenerative disorders. Accumulating evidence suggests that age-related mitochondrial impairment plays a fundamental role in the pathogenesis of Alzheimer's disease (AD), such as insufficient energy supply and excessive ROS production. Therefore, improving mitochondrial function may simultaneously target multiple AD pathologies. One option is to take advantage of the intrinsic mitochondrial quality control pathways that have evolved to identify and tackle the dysfunctional mitochondria and restore the health of organisms. Here we report that a non-antibacterial tetracycline derivative, 9-tert-butyl

tetracycline (9-TB), showed therapeutic effects in animal models of AD through activating the one of the mitochondrial quality control pathways - the mitochondrial stress response (MSR). In *Caenorhabditis elegans* models of AD, modulating MSR ameliorated amyloid proteotoxicity. Similarly, in a mouse model of AD, restoring mitochondrial homeostasis mitigated cognitive decline and electrocorticography abnormalities without affecting the gut microbiota. Ongoing investigations into the underlying mechanisms of action, such as ROS generation, neuronal loss and neuroinflammation will reveal how the improved mitochondrial population benefits brain function thus paving the way for the development of mitochondria-targeting therapies for AD.

Disclosures: Q. Wang: None. **S. Rodríguez-Lopez:** None. **T.Y. Li:** None. **J. Morel:** None. **M. Nelson:** None. **J. Auwerx:** None.

Poster

PSTR133. Parkinson's Disease: LRRK2 Mechanisms, Targets, and Pathways

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR133.01/E38

Topic: C.03. Parkinson’s Disease

Support: NIH R01 NS110879-05
Michael J. Fox Foundation for Parkinson’s Research grant #010325

Title: Serum urate correlations with urine exosome LRRK2 in early Parkinson disease.

Authors: *R. BAKSHI[1], Y. YUAN[2], A. CHANG[3], E. MACKLIN[4], A. B. WEST[2], M. A. SCHWARZSCHILD[1];
[1]Neurol., Massachusetts Gen. Hosp., Boston, MA; [2]Pharmacol., Duke Univ., Durham, NC; [3]Univ. of Minnesota, Minneapolis, MN; [4]Biostatistics Center, Massachusetts Gen. Hosp., Boston, MA

Abstract: Title: Serum urate correlations with urine exosome LRRK2 in early Parkinson disease.
Authors: Rachit Bakshi, Yuan Yuan, Allison Chang, Eric Macklin, Andrew West, and Michael Schwarzschild.
Higher serum urate is associated with a lower incidence and slower progression of PD and has been identified as a biomarker of resistance to PD among *LRRK2* mutation carriers. The SURE-PD (Safety of URate Elevation in Parkinson’s Disease) study was a Phase 2, randomized, double-blind, placebo-controlled trial of oral inosine to assess the safety of elevating serum urate in early PD patients with lower serum urate levels at baseline. Biofluids including urine were collected serially for PD-related biomarkers studies. Total LRRK2 protein was measured from urine extracellular vesicle fractions from biobanked SURE-PD samples at baseline, six months, and ~18 months in placebo (serum urate < 5.8mg/dL, n=23), mild (target serum urate 6.1-7.0 mg/dL, n=24) and moderate (target serum urate 7.1-8.0 mg/dL, n=24) groups. Biomarker and serum urate levels were compared at baseline by simple correlation. Association between change

in total LRRK2 levels and increase in serum urate was tested in a mixed model adjusting for sex, age, baseline serum urate, and baseline LRRK2 level. At baseline higher serum urate correlated with lower urinary LRRK2 levels (normalized to urine creatinine) *(r=-0.297, p = 0.013)* with a significant correlation in men (r= -0.428, *p=0.015*) but not women (r= -0.137, *p=0.41*). Change in serum urate from pre-treatment to post-treatment was also inversely correlated with change in urinary LRRK2 among men (-25% per mg/dL increase, *p=0.04*), women (-20% per mg/dL increase, *p=.03*), and overall (-22% per mg/dL increase, 95% CI -8% to -34%, *p=0.005*). These findings identify a possible association between serum urate and urine LRRK2 levels in early PD. Comparable analyses in plasma and CSF are ongoing to explore the extent of the association across compartments.

Disclosures: R. Bakshi: None. **Y. Yuan:** None. **A. Chang:** None. **E. Macklin:** None. **A.B. West:** None. **M.A. Schwarzschild:** None.

Poster

PSTR133. Parkinson's Disease: LRRK2 Mechanisms, Targets, and Pathways

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR133.02/E39

Topic: C.03. Parkinson’s Disease

Title: Decoding Parkinson's Disease Pathology: Spatial Phenotyping through Highly Multiplexed Imaging

Authors: J. NICHOLS[1], S. BUKHARI[2], K. AKABI[2], M. SCHONEMANN[2], M. E. MURRAY[3], A. PRATAPA[4], S. MISTRY[4], ***N. MAMMADOVA**[4], D. W. DICKSON[3], B. SCHUELE[2], T. J. MONTINE[2];
[1]St. Jude Children's Hosp., Memphis, TN; [2]Stanford Univ., Stanford, CA; [3]Mayo Clin., Jacksonville, FL; [4]Akoya Biosci., Menlo Park, CA

Abstract: Parkinson's disease (PD), a complex neurodegenerative disorder, poses a significant challenge in unraveling its complete pathology due to the intricate interplay of cellular and pathological manifestations. While neuropathological examinations of post-mortem brain samples have uncovered notable characteristics such as the loss of nigral neurons and the presence of alpha-synuclein inclusions, alongside markers of inflammation and tau deposition, the comprehensive understanding of this complex disease remains elusive. Techniques such as single-cell sequencing have the power to detect highly complex expression signatures from an individual cell but at the cost of spatial resolution at the cellular and subcellular levels. This warrants the need for protein level correlates that can capture both spatial and single-cell level measurements. To provide a detailed landscape of the pathology observed in PD, we established a panel of oligo-labeled antibodies for highly multiplexed imaging of human post-mortem PD samples using Akoya Biosciences PhenoCycler-Fusion technology. Our panel of 30+ proteins include markers of neuronal cells, immune cells, epithelial cells, pathological markers (Synuclein, TDP-43, Tau, and amyloid beta) and a marker of LRRK2 signaling (Rab12

pSer112). We applied our antibody panel to a cohort of samples of substantia nigra, striatum and nucleus basalis from non-neurologically compromised controls, age matched PD and LRRK2-PD autopsy samples. For each sample, we performed cell segmentation and average intensity computation followed by unsupervised clustering to characterize major cell phenotypes and their spatial distribution. We present a panel of markers for highly multiplexed characterization of PD phenotypes that quantitatively distinguishes between healthy control and PD autopsy samples.

Disclosures: J. Nichols: None. **S. Bukhari:** None. **K. Akabi:** None. **M. Schonemann:** None. **M.E. Murray:** None. **A. Pratapa:** None. **S. Mistry:** None. **N. Mammadova:** None. **D.W. Dickson:** None. **B. Schuele:** None. **T.J. Montine:** None.

Poster

PSTR133. Parkinson's Disease: LRRK2 Mechanisms, Targets, and Pathways

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR133.03/E40

Topic: C.03. Parkinson’s Disease

Support: the Grants-in-Aid for Scientific Research (A) 19H01015
the Grants-in-Aid for Scientific Research (C) 17K08265, 21K06558
Biogen

Title: Regulation of intracellular lysosomal positioning by LRRK2

Authors: *K. ITO[1], M. ARAKI[1], Y. KATAI[2], Y. NISHIMURA[2], S. IMOTANI[2], H. INOUE[2], G. ITO[3,4], T. TOMITA[1,4];
[1]Lab. of Neuropathology and Neuroscience, Grad. Sch. of Pharmaceut. Sciences, The Univ. of Tokyo, Tokyo, Japan; [2]HACARUS, Inc., Kyoto, Japan; [3]Dept. of Biomolecular Chemistry, Fac. of Pharma-Science, Teikyo Univ., Tokyo, Japan; [4]Social Cooperation Program of Brain and Neurolog. Disorders, Grad. Sch. of Pharmaceut. Sciences, The Univ. of Tokyo, Tokyo, Japan

Abstract: Parkinson disease (PD) is the second most common neurodegenerative disease, characterized by late-onset motor symptoms such as resting tremor. Progressive degeneration of dopaminergic neurons in the substantia nigra is thought to be the cause of PD symptoms. *Leucine-rich repeat kinase 2 (LRRK2)* has been identified as a causative gene for familial PD and as a locus associated with an increased risk of developing sporadic PD. LRRK2 physiologically phosphorylates Rab proteins involved in intracellular vesicle trafficking. Previous studies have shown that all FPD-linked mutations in LRRK2 abnormally increase the levels of Rab phosphorylation in cells. Based on these findings, it has been hypothesized that overactivation of LRRK2 causes neurodegeneration in PD. However, the exact mechanism is still unknown. In the present study, we found that overexpression of LRRK2 harboring an FPD mutation caused perinuclear clustering of lysosomes in cultured cells. The clustering was not observed when cells were treated with a specific LRRK2 inhibitor, suggesting that overactivation of LRRK2 is the cause of perinuclear clustering of lysosomes. Furthermore, perinuclear

clustering was abolished by knocking out Rab12 or Rab-interacting lysosomal protein-like 1 (RILPL1), an effector protein of Rab12. Re-expression of Rab12 in Rab12 knockout cells demonstrated that phosphorylation at Ser106 of Rab12 was required for perinuclear clustering of lysosomes. Finally, we showed that the interaction between Rab12 and RILPL1 was enhanced by phosphorylation of Rab12.Taken together, these results suggest that LRRK2 overactivation impairs proper lysosomal positioning in a Rab12-RILPL1-dependent manner, leading to neurodegeneration in PD.

Disclosures: K. Ito: None. **M. Araki:** None. **Y. Katai:** None. **Y. Nishimura:** None. **S. Imotani:** None. **H. Inoue:** None. **G. Ito:** None. **T. Tomita:** None.

Poster

PSTR133. Parkinson's Disease: LRRK2 Mechanisms, Targets, and Pathways

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR133.04/E41

Topic: C.03. Parkinson’s Disease

Support: Innovation Fund Denmark

Title: LRRK2 inhibition modulates cell death induced by lysosomal stressors in the mouse macrophage-like RAW 264.7 cell line

Authors: *J. TENGBERG[1,2], T. BENNED-JENSEN[1], J. NIELSEN[1];
[1]Mol. and single cell pharmacology, H Lundbeck, Copenhagen, Denmark; [2]Drug design and pharmacology, Univ. of Copenhagen, Copenhagen, Denmark

Abstract: Activating mutations in Leucine Rich Repeat Kinase 2 (LRRK2) strongly increase the risk for developing Parkinson’s disease (PD). The mechanistic path from LRRK2 mutations to PD is not established, but LRRK2 modulation of lysosomal function appears likely to be involved. Accumulating evidence indicates that regulation of lysosomal trafficking is an important function of LRRK2, and genes enriched in lysosomal function including LRRK2 are enriched in PD genome-wide association studies (Chang et al. 2017). Recent literature (Eguchi et al. 2018, Herbst et al. 2020, Bonet-Ponce et al. 2020) has reported that lysosomal damage induced by lysosomotropic agents such as LLOMe and chloroquine can activate LRRK2, which we have replicated. To determine if LRRK2 activity can conversely modulate the impact of lysosomotropic agents, we assessed the effect of LRRK2 inhibition on LLOMe-induced cell death in the macrophage-like mouse cell RAW 264.7, which has high endogenous LRRK2 expression. We found that the selective LRRK2 kinase inhibitors concentration-dependently attenuated LLOMe-induced RAW 264.7 cell death measured with a variety of confocal imaging and microplate-reader-based assays. The effect appears lysosomal specific as LRRK2 inhibition did not affect cell death induced by a variety of other cell death inducers that did not directly affect lysosomes. In addition, data on the mechanisms underlying LRRK2 inhibitor modulation

of lysosomal damage will be shown - including lysotracker-based measurement of lysosomal integrity, RNAseq analysis of transcriptional footprints, and knockdown of key genes.

Disclosures: **J. Tengberg:** A. Employment/Salary (full or part-time); H. Lundbeck A/S. **T. Benned-Jensen:** A. Employment/Salary (full or part-time); H. Lundbeck A/S. **J. Nielsen:** A. Employment/Salary (full or part-time); H. Lundbeck A/S.

Poster

PSTR133. Parkinson's Disease: LRRK2 Mechanisms, Targets, and Pathways

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR133.05/F1

Topic: C.03. Parkinson’s Disease

Support: Aligning Science Across Parkinson's (ASAP)
Queen Elizabeth II Graduate Scholarship
University of Ottawa

Title: Investigating the impact of Parkinson’s disease-associated mutation, Lrrk2^{G2019S}, on innate immunity

Authors: *K. HURLEY, M. SCHLOSSMACHER, S. SAD;
Univ. of Ottawa, Ottawa, ON, Canada

Abstract: The global prevalence of Parkinson’s disease (PD) is increasing faster than any other neurodegenerative disease, yet the cause of PD remains unknown. Recent studies indicate that alterations in innate immunity and chronic inflammation may play a role in the development and progression of PD. One of the common genetic mutations associated with PD affects a gene encoding leucine-rich repeat kinase 2 (Lrrk2). Interestingly, Lrrk2 expression is relatively low in the brain regions affected by PD but is abundantly expressed in immune cells and tissues. Furthermore, Lrrk2 has been associated with other immune system-related diseases including Crohn’s disease and leprosy. We propose that an agonistic mutation of Lrrk2, Lrrk2^{G2019S}, results in an enhanced immune response, and that this dysregulation of the immune response contributes to neuroinflammation and neurodegeneration in PD. Here, we use mice carrying the knock-in Lrrk2^{G2019S} mutation to study the impact of this mutation on the bone marrow (BM) and peripheral immune compartments. To understand the role of Lrrk2 in innate immunity, adult mice were intravenously inoculated with *Salmonella typhimurium* (ST), resulting in sepsis. Using flow cytometry, we determined that Lrrk2 expression is significantly upregulated in hematopoietic progenitor and mature myeloid cells following infection. Importantly, mice carrying the Lrrk2^{G2019S} mutation effectively controlled infection of the BM better than wildtype mice, indicated by a reduction in the number of ST colony forming units in the BM. To further determine which BM cell types were most responsible for the Lrrk2-mediated protection, we isolated bone marrow-derived macrophages (BMDMs) and neutrophils from adult mice. Following *in vitro* infection with ST, both BMDMs and neutrophils harbouring the agonistic

Lrrk2 mutation demonstrated a significant reduction in bacterial burden, relative to wildtype cells. As well, to better understand the function of Lrrk2 in the BM compartment, wildtype adult mice were subjected to lethal doses of irradiation and subsequently transplanted with a fifty-fifty mixture of wildtype and $Lrrk2^{G2019S}$ bone marrow cells. Flow cytometry results indicate that immune cells harbouring the $Lrrk2^{G2019S}$ mutation displayed a selective reconstitution advantage in comparison to wildtype cells, suggesting that Lrrk2 may promote an enhanced immune response through improved hematopoietic function. Principally, the results of this study will provide new insights into the understanding of how agonistic Lrrk2 mutations may contribute to PD pathology through an amplified immune response that ultimately contributes to neuroinflammation.

Disclosures: K. Hurley: None. **M. Schlossmacher:** None. **S. Sad:** None.

Poster

PSTR133. Parkinson's Disease: LRRK2 Mechanisms, Targets, and Pathways

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR133.06/F2

Topic: C.03. Parkinson’s Disease

Title: Peripheral immune system dysregulation in G2019S LRRK2 mice following infection with influenza induces neurodegenerative changes in the SNpc

Authors: *K. CROWTHER[1], E. KOZINA[3], D. CHATTERJEE[2], R. J. SMEYNE[2];
[2]Thomas Jefferson Univ., [1]Farber Inst. of Neurosci. Thomas Jefferson Univ., Philadelphia, PA;
[3]Thomas Jefferson Univ., Thomas Jefferson Univ., Philadelphia, PA

Abstract: Parkinson’s Disease (PD) is the 2nd most common neurodegenerative disease, affecting 1 million patients in the US. Clinically, it is characterized by a myriad of both motor and non-motor symptoms. The characteristic neuropathological changes in PD involve loss of the DA neurons in the SNpc and the presence of Lewy bodies that primarily consist of insoluble aggregates of -synuclein. In addition to these CNS changes, PD patients also demonstrate a clear dysregulation in the immune system, both centrally and peripherally. While the pathology in PD is understood, its exact etiology has long been debated. Current theories posit a role for environment, infection, and genes in PD etiology. About 15% of all PD cases are linked to a known genetic risk factor. A common PD mutation occurs at position 2019 in the LRRK2 gene (G2019S). LRRK2 functions as a kinase and has its highest expression in immune cells, lungs, and brain. The G2019S LRRK2 mutation enhances kinase activity, which has a multitude of downstream effects including increased activity of the immune system. However, LRRK2’s penetrance is about 25%, suggesting that an additional trigger is needed to provoke its pathogenicity. The genetic and environmental interaction is known as the multi-hit hypothesis of PD. One environmental risk factor is viral infection, including influenza. Historically, there have been several influenza pandemics associated with significant outbreaks of neurological manifestations, including alterations in cognition, encephalitis, and acute parkinsonism. In this

study, we sought to determine if the 2009 pandemic influenza virus (H1N1, strain A/CA/04/2009) was capable of synergizing with mutant LRRK2 to increase development of parkinsonian pathology. In 12-month uninfected LRRK2 G2019S mice, no differences were seen in SNpc DA neuron number. Additionally, in H1N1 infected WT mice we saw no neuropathology. However, in H1N1-infected G2019S mice, we found a 20% cell loss starting at 7d post infection. Further, infected-G2019S mice also showed a significant decrease in survivability compared with infected-WT animals. Examination of the brain during this period of infection did not show any evidence for intraparenchymal viral proteins or T-cell infiltration. The lack of direct viral infection or immune cell invasion suggests a critical importance of the signals that emanate from the peripheral immune system in the initiation of the neuropathological process. Further studies are currently ongoing to characterize both the peripheral and CNS immune profiles to better define the role of dysregulated LRRK2 activity secondary to viral infection during the development of experimental PD.

Disclosures: **K. Crowther:** None. **E. Kozina:** None. **D. Chatterjee:** None. **R.J. Smeyne:** None.

Poster

PSTR133. Parkinson's Disease: LRRK2 Mechanisms, Targets, and Pathways

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR133.07/F3

Topic: C.03. Parkinson’s Disease

Support: #NEXTGENERATIONEU (NGEU) and funded by the Ministry of University and Research (MUR), National Recovery and Resilience Plan (NRRP), project MNESYS (PE0000006)

Title: Enhanced D1 dopaminergic tranmsission in aged LRRK2 G2019S knock-in mice

Authors: D. MERCATELLI[1,2], A. BRUGNOLI[1], F. ALBANESE[1], A. DI MAIO[3], D. R. SHIMSHEK[4], A. USIELLO[3], ***M. MORARI**[1];
[1]Neurosci. and Rehabil., Univ. of Ferrara, Ferrara, Italy; [2]LTTA Lab. for Advanced Therapies, Technopole of Ferrara, Italy; [3]Univ. Vanvitelli, Univ. della Campania, Caserta, Italy; [4]Novartis Pharma AG, Novartis Pharma AG, Basel, Switzerland

Abstract: LRRK2 G2019S knock-in (KI) mice represent a valuable model to study prodromic changes of basal ganglia transmission in Parkinson’s disease. Here, we performed neurochemical, biochemical and behavioral analysis in 3-month-old and 12-month-old G2019S KI and wild-type (WT) mice to investigate whether the G2019S mutation is associated with enhanced D1 dopaminergic transmission over aging. Dual probe intracerebral microdialysis revealed elevations of striatal and nigral glutamate levels and reduction of nigral GABA levels in 12-month-old G2019S KI mice. Reverse dialysis of the D1 receptor antagonist SCH-23390 in striatum selectively elevated striatal GABA release in 12-month-old G2019S KI mice.

Intrastriatal SCH-233390 was associated with a prolonged reduction of glutamate release in the substantia nigra reticulata of G2019S KI and WT mice along with no changes of nigral GABA release. Systemic administration of the D1 receptor agonist SKF-81297 did not affect neurotransmitter release in any genotypes. Likewise, Western blotting revealed similar increases of striatal phosphorylated extracellular signal-regulated kinase 1 and 2 (pERK) and Glutamate Receptor 1 (pGluA1) in 3-month-old G2019S KI and WT mice challenged with SKF-81297. Conversely, pGluR1 levels were elevated by SKF-81297 to a greater extent in 12-month-old G2019S KI mice compared to age-matched controls. Behavioral analysis showed elevated grooming activity in 12-month-old G2019S KI mice compared to age-matched WT and LRRK2 kinase-dead mice. Moreover, 12-month-old G2019S KI mice exhibited a more prolonged hypokinetic response to systemic SCH-23390 compared to wild-type mice. We conclude that G2019S KI mice show age-dependent enhancement of endogenous D1 transmission in the striatum.

Disclosures: D. Mercatelli: None. **A. Brugnoli:** None. **F. Albanese:** None. **A. Di Maio:** None. **D.R. Shimshek:** A. Employment/Salary (full or part-time); Novartis Pharma. **A. Usiello:** None. **M. Morari:** None.

Poster

PSTR133. Parkinson's Disease: LRRK2 Mechanisms, Targets, and Pathways

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR133.08/F4

Topic: C.03. Parkinson’s Disease

Support: NIH-R01NS120879 to WWS
NIH-R01NS119208 to WWS

Title: Neurovascular abnormalities in a G2019S-LRRK2 mouse model of Parkinson disease

Authors: H. FUEHRER[1], K. KURREY[1], B. NING[1], L. LIN[1], M. MERCADO GUERRA[1], M. JIANG[1], A. BIBIC[1,2], P. VAN ZIJL[1,2], C. ROSS[1], J. HUA[1,2], ***W. SMITH**[1];
[1]John Hopkins Univ. Sch. of Med., Baltimore, MD; [2]Kennedy Krieger Inst., Baltimore, MD

Abstract: Parkinson's disease (PD) is a common neurodegenerative disease characterized by motor impairments resulting from midbrain dopamine (DA) neuron loss. Mutations in LRRK2 cause genetic PD and contribute to sporadic PD. Here, we used G2019S-LRRK2-transgenic mouse model to investigate abnormalities in arteriolar cerebral blood volume (CBVa) and lymphatic vessels in various brain regions using the inflow-based vascular-space-occupancy (iVASO) and gadolinium-based MRI techniques. CBVa measured brain regions included in the substantia nigra (SN, the PD affected area), olfactory cortex and prefrontal cortex. Alterations in the blood volume of small arteries and arterioles (CBVa) were detected in the G2019S-LRRK2 mouse model of PD. Compared to non-transgenic mice, G2019S-LRRK2 mice at preclinical clinical stage showed increased CBVa and at clinical stage showed decreased CBVa in the SN in

both male and female groups. On contrast, WT-LRRK2 mice showed no change in CBVa in the SN (male and female). Moreover, the G2019S-LRRK2 also induced abnormal dynamic signal changes of lymphatic vessels in the basal region (BR) compared with non-transgenic mice at both preclinical and clinical stages. These MRI changes in G2019S-LRRK2 mice was validated by pathological studies and was corresponding with PD pathology. Our results suggest the brain MRI changes of CBVa and lymphatic vessels may be useful as a marker for PD disease progression.

Disclosures: H. Fuehrer: None. **K. Kurrey:** None. **B. Ning:** None. **L. Lin:** None. **M. Mercado Guerra:** None. **M. Jiang:** None. **A. Bibic:** None. **P. Van Zijl:** None. **C. Ross:** None. **J. Hua:** None. **W. Smith:** None.

Poster

PSTR133. Parkinson's Disease: LRRK2 Mechanisms, Targets, and Pathways

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR133.09/F5

Topic: C.03. Parkinson’s Disease

Title: Characterization of LRRK2 Pathogenic Signaling through Small GTPase Rab Protein Interactions in Parkinson’s Disease

Authors: *X. LI;
Icahn Sch. of Med. at Mount Sinai, New York, NY

Abstract: Characterization of LRRK2 Pathogenic Signaling through Small GTPase Rab Protein Interactions in Parkinson’s Disease
Xingjian Li[1], Hanwen Zhu[2], Insup Choi[1], Bik Tzu Huang[1], Dongxiao Liang[1], Xianting Li[1], Ji Sun[2], Zhenyu Yue[1]*1 Department of Neurology and Department of Neuroscience, The Friedman Brain Institute, Icahn School of Medicine at Mount Sinai, New York, NY, USA2 Department of Structural Biology, St. Jude Children's Research Hospital, Memphis, TN, USA
Parkinson’s disease (PD) is the second most prevalent neurodegenerative disease. The exact pathogenic mechanisms of PD are poorly understood. LRRK2 variants have been linked to the most common familial forms of PD. LRRK2 was shown to regulate various cellular pathways, including autophagy-lysosome, synaptic vesicle trafficking, mitochondria homeostasis, ciliogenesis, and immune response. In addition, multiple lines of evidence demonstrated that LRRK2 phosphorylates a subset of Rab GTPases in vitro and in vivo. But the structural and functional significance and consequence of LRRK2-mediated signaling through various Rab proteins remains to be clarified. We have investigated the structure of a specific LRRK2-Rab complex and characterized their functional interaction in ciliogenesis. Our results showed that a small GTPase Rab protein regulates ciliogenesis in glia and mediates LRRK2 mutant-associated cilia deficiency. Our study provides an insight into the molecular mechanism of LRRK2 signaling through concerted interactions with multiple Rab proteins, which may underlie the

pathogenesis of LRRK2-associated PD.
Keywords: LRRK2, Rab GTPases, Parkinson's Disease

Disclosures: X. Li: None.

Poster

PSTR133. Parkinson's Disease: LRRK2 Mechanisms, Targets, and Pathways

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR133.10/F6

Topic: C.03. Parkinson's Disease

Support: The Michael J. Fox Foundation (MJFF)

Title: Characterization of the autophagic-lysosomal pathway in parkinson's disease using patient ipsc-derived dopaminergic neurons containing lrrk2 or gba mutations

Authors: *S. COVENEY[1], K. LEFRANCOIS[1], V. BAIN[1], M. ZOLLER[1], S. SINGH[1], A. FATHI[2], S. SCHACHTELE[2], R. W. CHO[1];
[1]Cell Signaling Technology, Inc., Danvers, MA; [2]FUJIFILM Cell. Dynamics, Madison, WI

Abstract: Dopaminergic neuron cell death in Parkinson's Disease (PD) is complex and combinatorial, with impairments in multiple cellular pathways impacting mitochondrial function, endosomal/lysosomal protein degradation, alpha-synuclein and tau aggregation, and neuroinflammation. Genetic risk factors, such as mutations in leucine-rich repeat kinase 2 (LRRK2) and glucocerebrosidase (GBA), have been shown to impact kinetics of the autophagic-lysosomal pathway (ALP), which is suggested to contribute to PD-associated protein accumulation and aggregation. Human-relevant in vitro models using patient-derived induced pluripotent stem cells (iPSCs) offer an accessible avenue for understanding the mechanisms for these genetic mutations as well for development of therapeutics against this debilitating disease. In this study, we evaluated the expression and cellular distribution of ALP-associated proteins in iPSC-derived dopaminergic neurons (iCell® DopaNeurons) generated from both apparently healthy normal donors (AHN) and patients clinically diagnosed with Parkinson's Disease and harboring LRRK2 G2019S or GBA1 N370S mutations. Disease iPSCs were obtained from the Parkinson's Progression Markers Initiative (PPMI), part of The Michael J. Fox Foundation (MJFF). Using high-throughput imaging we quantified endosome and lysosome protein expression using s an array of highly specific antibodies from Cell Signaling Technology (CST), including LAMP-1, Rab5, LC3, DRP1, Cathepsin, and Galectin 3. As the onset of PD is multifactorial, we applied exogenous chemical modulators of autophagy (Torin 1) and mitochondrial stress (Rotenone) to the LRRK2, GBA and AHN iPSC-derived dopaminergic neurons to evaluate the effect of additional stressors on ALP protein expression. We also evaluated neural survival and network formation between AHN and the PD iPSC-derived dopaminergic neurons by quantifying tyrosine hydroxylase and synaptic markers (ie., synapsin 1, synaptophysin, PSD95, and VAMP2), respectively. These markers were used to quantify

neurodegeneration following exposure to Torin 1 or Rotenone. Together, these data demonstrate the utility of high-throughput immunocytochemistry and patient-derived iPSC dopaminergic neurons for investigating lysosomal, mitochondrial, neurodegenerative pathway dynamics.

Disclosures: **S. Coveney:** A. Employment/Salary (full or part-time); Cell Signaling Technology. **K. LeFrancois:** A. Employment/Salary (full or part-time); Cell Signaling Technology. **V. Bain:** A. Employment/Salary (full or part-time); Cell Signaling Technology. **M. Zoller:** A. Employment/Salary (full or part-time); Cell Signaling Technology. **S. Singh:** A. Employment/Salary (full or part-time); Cell Signaling Technology. **A. Fathi:** A. Employment/Salary (full or part-time); FUJIFILM Cellular Dynamics. **S. Schachtele:** A. Employment/Salary (full or part-time); FUJIFILM Cellular Dynamics. **R.W. Cho:** A. Employment/Salary (full or part-time); Cell Signaling Technology.

Poster

PSTR133. Parkinson's Disease: LRRK2 Mechanisms, Targets, and Pathways

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR133.11/F7

Topic: C.03. Parkinson’s Disease

Support: NIH R01 NS112506
NIH K01 AG046366
Parkinson’s Foundation Stanley Fahn Junior Faculty award PF-JFA-1934
UConn Startup fund
NIH R01 GM136904
NSF 2115690

Title: Regulation of LRRK2 mRNA stability by ATIC and its substrate AICAR through ARE-mediated mRNA decay in Parkinsons disease

Authors: Q. LIU[1], D. ZHU[1], N. LI[2], S. CHEN[1], L. HU[2], J. YU[2], ***Y. XIONG**[1];
[1]Neurosci., Univ. of Connecticut Sch. of Med., Farmington, CT; [2]Physiol. & Neurobio., Univ. of Connecticut, Storrs, CT

Abstract: Mutations in LRRK2 are the most common genetic causes of Parkinson’s disease (PD). Increasing studies revealed elevated LRRK2 protein expression in certain PD patient tissues without LRRK2 enzymatic dysfunctions, suggesting an important role of LRRK2 protein levels in PD pathogenesis. The mechanism underlying the regulation of LRRK2 protein levels remains unclear. Here, we identified ATIC as a novel regulator that regulates LRRK2 levels and toxicity. We discovered that AICAr, the precursor of ATIC enzymatic substrate AICAR/ZMP, mediates LRRK2 levels in vitro through a cell-type specific regulation and in vivo in mouse tissues. Mechanistically, AICAr regulates LRRK2 levels through AUF1-mediated mRNA decay. Upon AICAr treatment, AUF1 is recruited to the AU-rich elements (ARE) of LRRK2 mRNA, and the decapping enzyme complex DCP1/2 is promoted, leading to the decay of LRRK2

mRNA. Importantly, we found that AICAr suppresses LRRK2 expression and rescues LRRK2-induced dopaminergic neurodegeneration and neuroinflammation in PD animal models. Together, this study provides the first and a novel regulatory mechanism of LRRK2 function through LRRK2 mRNA decay, which is distinct from LRRK2 enzymatic functions.

Disclosures: **Q. Liu:** None. **D. Zhu:** None. **N. Li:** None. **S. Chen:** None. **L. Hu:** None. **J. Yu:** None. **Y. Xiong:** None.

Poster

PSTR134. Parkinson's Disease: Small Molecule Therapeutics

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR134.01/F8

Topic: I.08. Methods to Modulate Neural Activity

Support: RF1-MH117055
WU-22-0377
ASAP-020607

Title: Studying anxiety-like behavior in mice using cell type-specific diazepam

Authors: ***A. S. MIN**[1], H. YAN[2], B. C. SHIELDS[2], M. R. TADROSS[2];
[1]Neurobio., [2]Biomed. Engin., Duke, Durham, NC

Abstract: Anxiety disorders are the most prevalent psychiatric condition and a leading cause of disability. Treatment involves psychotherapy and medication, yet only 60-85% of patients respond. Despite a clear need for novel medications, drug development has shifted away from anxiety disorders in the past decade. To deliver better outcomes to patients, an improved understanding of the neurobiological basis of anxiety is necessary. While the primary drug-receptor interactions and resultant behavioral profiles are well characterized, the circuit mechanisms through which molecular events transduce to behavioral changes remains a daunting mystery. Commendable advancements have been made in mapping anxiety-relevant neural circuits using chemo- and optogenetics. By manipulating activity in precise cell types and brain regions, these approaches have emphasized the importance of cell types in anxiety-like behavior. They inform a circuit understanding, but one complicated by overexpression of exogenous actuators and not concerned with pharmacology. To overcome these limitations, it was necessary to develop DART (drugs acutely restricted by tethering). With DART we can genetically program a subset of cells to capture and concentrate a specific drug to levels ~1000 times higher than anywhere else. Therapeutic doses of a clinical drug can be sequestered to a specific cell, without effects on neighboring neurons or axons of passage. We selected diazepam.1[DART.2] to study anxiety because diazepam exhibits an anxiolytic effect time-locked to the concentration of drug in the brain, indicating that it acts on a proximate causal determinant of anxiety. This contrasts with other first line pharmacotherapies such as SSRIs which exhibit a complex and delayed response. Diazepam is a benzodiazepine (BZD), a class of positive allosteric modulators

(PAM) of the $GABA_AR$. Unlike other cell-type specific manipulations, diazepam.1$^{DART.2}$ shapes activity through endogenous receptors and without imposing any external pattern of input. This affords an unprecedented opportunity to bridge the gap between clinical pharmaceuticals and cell type-specific circuit-deconstruction paradigms in neuroscience. With this tool we are identifying which brain regions and cell-types are sufficient for diazepam's anxiolytic effect on the elevated plus maze (EPM) and demonstrating that on-target effects can be disentangled from off-target effects such as locomotion. These studies will inform the development of next generation medications for anxiety with fully understood mechanisms bridging molecular action through circuit to behavior.

Disclosures: A.S. Min: None. **H. Yan:** None. **B.C. Shields:** None. **M.R. Tadross:** None.

Poster

PSTR134. Parkinson's Disease: Small Molecule Therapeutics

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR134.02/G1

Topic: I.08. Methods to Modulate Neural Activity

Support: R61DA051530

Title: Deconstructing the neural correlates of opioid use disorder with the opioid-DART toolset

Authors: *S. YOUSEFZADEH[1], H. YAN[3], B. C. SHIELDS[2], P. JEONG[2], Y. OH[2], S. KWAK[4], J. HONG[2], M. TADROSS[2];
[1]Psychology and Neurosci., [2]Duke Univ., Durham, NC; [3]Dept. of Neurobio., Duke university Sch. of Medicine, Dept. of neurobiology, Durham, NC; [4]Ambagon therapeutics, San Francisco, CA

Abstract: In the search for neurobiological underpinnings of opioid addiction, disentangling the analgesic and addictive properties of opioid compounds has been elusive, since both properties are carried out through the same opioid receptors expressed in distinct cell types. In classical pharmacology, drugs exert their effects on target receptors in all cells within a given brain volume, which may lead to various downstream responses from different cell-types that are not dissociable from each other. In order to circumvent this issue, our lab developed "Drug Acutely Restricted by Tethering (DART)" to allow cell-type specific pharmacological manipulations. This technology relies on expression of the HaloTag protein (HTP) in the cell-type of interest followed by administration of a drug conjugated with the HaloTag ligand (HTL). This configuration covalently binds and restricts the action of the drug to the defined cell type, while having negligible ambient effects. By extending this technology to opioid drugs, we have established an opioid-DART toolset to study the neural correlates of opioid addiction. Furthermore, we have characterized these compounds through the Tango GPCR assay system to quantify their efficacy and potency, validated their ability to produce cell-specific effects on HTP+ neurons in brain slice, and are now applying the reagents to behaving mice. As a first step,

we are focusing on the dopamine-independent component of opioid addiction by targeting the local cholinergic interneurons in the nucleus accumbens. These cell-specific opioid manipulations offer unprecedented opportunities to uncover cellular and circuit contributions of opioid-mediated behaviors, with implications for the development of safer analgesics and rehabilitation strategies.

Disclosures: S. Yousefzadeh: None. **H. Yan:** None. **B.C. Shields:** None. **P. Jeong:** None. **Y. Oh:** None. **S. kwak:** None. **J. Hong:** None. **M. Tadross:** None.

Poster

PSTR134. Parkinson's Disease: Small Molecule Therapeutics

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR134.03/G2

Topic: I.08. Methods to Modulate Neural Activity

Support: NIH Grant R61DA051530
NIH Grant RF1-MH117055

Title: DART and a deeper understanding of catalepsy

Authors: *J. R. RAVENEL[1], S. YOUSEFZADEH[2], H. YAN[3], R. CARTER[1], B. C. SHIELDS[3], M. TADROSS[3];
[1]Neurobio., [2]Psychology and Neurosci., [3]Biomed. Engin., Duke Univ., Durham, NC

Abstract: Haloperidol and related antipsychotics have been central in the treatment of psychosis for more than 50 years, but their utility is severely hindered by Parkinsonian-like motor side effects that are referred to as extrapyramidal symptoms (EPS). In mice, haloperidol induces catalepsy, which is defined as an inability to correct an externally imposed posture and is measured with the bar-test. This assay has been a valuable predictive model for screening new generations of antipsychotics for EPS and provides a platform for examining circuit mechanisms. While it is known that antipsychotics primarily antagonize the D2 dopamine receptor (D2R), it has been difficult to disentangle circuit mechanisms owing to broad D2R expression in diverse cell types. To address this issue, we built upon the second-generation DART (Drug Acutely Restricted by Tethering) platform, which enables delivery of drugs to genetically defined cells, with little or no impact on neighboring neurons or axons of passage. We synthesized several haloperidol$^{DART.2}$ compounds, performed initial screening of these compounds using TANGO GPCR assays, and validated these compounds in slice electrophysiology, performing whole-cell patch clamp recordings of cholinergic interneurons in the dorsal striatum. With this toolset in hand, we proceeded to deliver haloperidol$^{DART.2}$ to cholinergic interneurons in awake mice, coupling it with the bar test, 3-dimensional behavior tracking, and fiber photometry. These studies are well positioned to reveal fundamental circuit mechanisms of antipsychotic-induced catalepsy, with implications for development of safer, more effective therapeutics.

Disclosures: **J.R. Ravenel:** None. **S. Yousefzadeh:** None. **H. Yan:** None. **R. Carter:** None. **B.C. Shields:** None. **M. Tadross:** None.

Poster

PSTR134. Parkinson's Disease: Small Molecule Therapeutics

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR134.04/G3

Topic: I.08. Methods to Modulate Neural Activity

Support: NIH 1RF1MH117055
NIH 1DP2MH119425
NIH 5R01NS107472
NIH 5R61DA051530

Title: DART.2—Optimized capture chemistry for cell-specific pharmacology

Authors: ***M. TADROSS**[1], B. C. SHIELDS[1], H. YAN[1], S. S. X. LIM[1], S. C. BURWELL[1], E. A. FLEMING[1], C. M. CAMMARATA[1], E. W. KAHUNO[1], P. VAGADIA[2], M. H. LOUGHRAN[3], L. ZHIQUAN[2], M. E. MCDONNELL[3], M. L. SCALABRINO[1], M. THAPA[1], T. M. HAWLEY[1], A. B. REITZ[3], G. E. SCHILTZ[2], C. HULL[1], G. D. FIELD[1], L. L. GLICKFELD[1];
[1]Duke Univ., Durham, NC; [2]Northwestern Univ., Evanston, IL; [3]FCCDC, Philadelphia, PA

Abstract: Neuropharmaceuticals can attenuate or allosterically strengthen chemical communication between brain cells. Cell-specific versions of these drugs hold enormous potential for causal interrogation of the connectome. In particular, **DART** (drug acutely restricted by tethering) genetically programs cells to accumulate tethered drug to levels higher than ambient concentrations. The technology achieves cellular specificity by instructing cells to express the HaloTag Protein (HTP), which captures and locally accumulates a drug linked to the HaloTag Ligand (HTL). Here we describe the DART.2 platform, comprising three advances. First, we optimized chemical attributes to achieve 3,000-fold cellular specificity, enabling safe delivery of an otherwise epileptogenic drug to genetically defined cells. Second, we achieved brain-wide dosing, without a local cannula, and developed reagents to visualize quantitative target engagement of drug capture in each animal. Third, we extended the method to enable either antagonism or positive-allosteric modulation of excitatory (AMPAR) or inhibitory (GABAAR) synapses. We validated the tools in ex-vivo slice, as well as in behaving mice. **DART.2** empowers a new kind of synaptic interrogation—between a genetically defined postsynaptic cell type, and chemically defined population of presynaptic cells. The approach offers design simplicity for ease-of-use in the neuroscience field, further facilitated via **DARTpharm**, a distribution platform for viral and chemical reagents.

Disclosures: **M. Tadross:** None. **B.C. Shields:** None. **H. Yan:** None. **S.S.X. Lim:** None. **S.C. Burwell:** None. **E.A. Fleming:** None. **C.M. Cammarata:** None. **E.W. Kahuno:** None. **P.**

Vagadia: None. **M.H. Loughran:** None. **L. Zhiquan:** None. **M.E. McDonnell:** None. **M.L. Scalabrino:** None. **M. Thapa:** None. **T.M. Hawley:** None. **A.B. Reitz:** None. **G.E. Schiltz:** None. **C. Hull:** None. **G.D. Field:** None. **L.L. Glickfeld:** None.

Poster

PSTR134. Parkinson's Disease: Small Molecule Therapeutics

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR134.05/G4

Topic: I.08. Methods to Modulate Neural Activity

Support: NIH Grant DP2MH1194025
FDN #ASAP-020607

Title: $Gaba_A$ receptors on dopamine neurons support conditioned conviction

Authors: *S. C. V. BURWELL[1], H. YAN[2], B. C. SHIELDS[1], S. YOUSEFZADEH[1], S. S. X. LIM[1], M. R. TADROSS[1];
[1]Duke Univ., Durham, NC; [2]Dept. of Neurobio., Duke university Sch. of Medicine, Dept. of neurobiology, Durham, NC

Abstract: Ventral tegmental area dopamine neurons (VTA_{DA}) fire in a manner consistent with Reward Prediction Error, with better-than-expected and worse-than-expected outcomes correlating with increases and decreases in firing, respectively. These dynamic firing patterns are induced by various neurochemical inputs, one of which is GABA, the main inhibitory neurotransmitter in the mammalian brain. Because GABA is released and sensed by many cell types, its specific role in regulating VTA_{DA} activity and the resulting impact on animal behavior remain unclear. Utilizing a novel tool, Drug Acutely Restricted by Tethering (DART), we were able to acutely block $GABA_A$ receptors exclusively on VTA_{DA} neurons, thereby diminishing their ability to receive inhibitory synaptic communication without affecting GABA release or its reception by surrounding cells. By pairing this tool with *in vivo* neural recordings, we found that this precise manipulation significantly reduces dynamic pauses in firing of VTA_{DA} neurons, a finding that aligns with the known role of $GABA_A$ receptors in synaptic inhibition. However, contrary to our hypothesis, performing this manipulation during an associative learning behavioral assay resulted in a twofold acceleration of extinction learning, causing animals to more rapidly abandon a cue once it was no longer associated with reward. Remarkably, the learning of a new cue-reward association remained unaffected in the same animals. These findings challenge previous assumptions about the role of $GABA_A$ communication to VTA_{DA} neurons, attributing it not to a pure extinction signal but instead to a conditioned conviction signal: a signal promoting the persistence of previously valuable associations despite counter-evidence to their current usefulness. This opens up new perspectives into the dynamic neurochemical regulation of behavioral persistence, with broad implications for our understanding of adaptive learning behaviors.

Disclosures: **S.C.V. Burwell:** None. **H. Yan:** None. **B.C. Shields:** None. **S. Yousefzadeh:** None. **S.S.X. Lim:** None. **M.R. Tadross:** None.

Poster

PSTR134. Parkinson's Disease: Small Molecule Therapeutics

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR134.06/G5

Topic: I.08. Methods to Modulate Neural Activity

Support: NIH Grant 1RF1MH117055-01
CBTE Lew Fellowship
AHA Predoctoral Fellowship

Title: Appraising the functional locality of receptors with subcellular pharmacology

Authors: *S.-X. LIM, S. SINGH, B. C. SHIELDS, S. C. BURWELL, A. CHOUDHURY, H. YAN, D. KOO, M. R. TADROSS;
Duke Univ., Durham, NC

Abstract: Neurons are highly polarized cells with distinct anatomical compartments (axon, soma, dendrites, primary cilia). These subcellular compartments are thought to serve distinct functions. However, it remains unknown how the same receptor in different subcellular compartments impacts circuit dynamics and animal behavior. A prototypical example is in regard to the main inhibitory neurotransmitter, GABA, whose receptor (the $GABA_AR$) is present throughout all locales of neurons. Each compartment is biophysically positioned to perform distinct functions, and receives distinct streams of information. For instance, $GABA_ARs$ on distal dendrites are thought to counterbalance to local excitatory *inputs*, whereas those on the soma are thought to have veto power over action potential *output*. $GABA_ARs$ in the axon may decouple transmitter release from action potential firing, with potential to individually tune collaterals. Beyond these traditional compartments, the primary cilia has recently been shown to be a distinct postsynaptic target, raising the tantalizing prospect for specialized $GABA_AR$ regulation of signaling to the nucleus. A second transmitter implicated in subcellular processing is dopamine, for which receptors exists in all these anatomical compartments. Dopamine receptors may have particular significance to the primary cilium, where dopamine receptors are enriched. DART (Drug Acutely Restricted by Tethering) offers a groundbreaking new way to study native receptors, including dopamine and $GABA_ARs$, by making it possible to deliver pharmaceuticals to genetically defined cells. The cells of interest are made to express a protein that can covalently capture and concentrate drugs to levels ~1,000-fold higher than the ambient concentration, yielding a localized cell-specific pharmaceutical effect. Here, we describe three novel subcellular refinements of the technology. First, we describe a method to deliver DART pharmaceuticals to axon projections without diffusion into somatodendritic compartments, enabling axon-specific pharmacology. Second, we present novel soma-targeted versions of DART. Finally, we describe

cilia-targeted versions of DART. These tools are compatible with use in freely behaving mice, and should provide a modular foundation for subcellular targeting of virtually any drug.

Disclosures: **S. Lim:** None. **S. Singh:** None. **B.C. Shields:** None. **S.C. Burwell:** None. **A. Choudhury:** None. **H. Yan:** None. **D. Koo:** None. **M.R. Tadross:** None.

Poster

PSTR134. Parkinson's Disease: Small Molecule Therapeutics

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR134.07/G7

Topic: I.08. Methods to Modulate Neural Activity

Support: DP2MH1194025
RF1MH117055

Title: Deconstructing the neural correlates of temporal lobe epilepsy with the cannabinoid-DART toolset

Authors: S. YOUSEFZADEH[1], K. R. JENSEN[3], ***B. SHIELDS**[1], S. S. LIM[1], M. MCDONNELL[4], A. B. REITZ[5], J. MAREK[1], J. O. MCNAMARA[2], M. TADROSS[1]; [2]Dept. of Neurobio., [1]Duke Univ., Durham, NC; [3]Albert Einstein Col. of Med., Bronx, NY; [4]Fox Chase Chem. Diversity Ctr., King of Prussia, PA; [5]Fox Chase Chem. Diversity Center, Inc., Doylestown, PA

Abstract: The type-I cannabinoid receptor (CB1R) is the most abundant receptor in the mammalian brain, and is particularly enriched in the dentate gyrus of the hippocampus, where it plays a key role in regulating circuit dynamics. CB1Rs are expressed in axon terminals of at least two distinct cell types, including the excitatory mossy cell (MC) and inhibitory cholecystokinin (CCK) cell. These cells provide synaptic input to dentate granule cells, which in turn generate endocannabinoids in an activity-dependent manner. As such, the CB1R is well positioned to dynamically regulate excitatory to inhibitory balance onto dentate granule cells. It has recently been shown that CB1Rs become upregulated in CCK cells during the progression of Temporal Lobe Epilepsy (TLE), a devastating disorder characterized by runaway excitation in the highly recurrent hippocampal circuitry. However, it has been difficult to determine whether this CB1R upregulation is compensatory or causal in the progression of TLE. To address this, we have developed new DART (Drug Acutely Restricted by Tethering) reagents that enable cell type-specific delivery of a CB1R agonist (THC.$1^{DART.2}$) or CB1R antagonist (rimonabant.$4^{DART.2}$). We have synthesized and validated these reagents using Tango GPCR assays and whole-cell patch clamp electrophysiology. Moreover, we have preliminary evidence that these CB1R DARTs can be exclusively delivered to CCK cells in awake mice to bidirectionally alter seizure thresholds in a kindling model of epilepsy. Our newly developed CB1R-DARTs provide a novel approach for manipulating endogenous CB1Rs in genetically defined cells, with broad relevance to endocannabinoid signaling in diverse neurological contexts and disease states.

Disclosures: **S. Yousefzadeh:** None. **K.R. Jensen:** None. **B. Shields:** None. **S.S. Lim:** None. **M. McDonnell:** None. **A.B. Reitz:** None. **J. Marek:** None. **J.O. McNamara:** None. **M. Tadross:** None.

Poster

PSTR134. Parkinson's Disease: Small Molecule Therapeutics

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR134.08/G8

Topic: I.08. Methods to Modulate Neural Activity

Support: NIH/Ro1NS107472

Title: Exploring Post-Synaptic Specificity of DART.2 in Excitatory Neurotransmission

Authors: *H. YAN[1], B. C. SHIELDS[1], S. S. X. LIM[1], S. BURWELL[2], M. TADROSS[1,2]; [1]Dept. of Biomed. Engin., Duke university, Durham, NC; [2]Dept. of neurobiology, Duke Univ., Durham, NC

Abstract: Drug Acutely Restricted by Tethering (DART) enables cell-specific drug delivery. The method works by expressing the HaloTag Protein (HTP) on genetically defined cells of interest, enabling these cells to capture and locally accumulate drugs linked to the HaloTag Ligand (HTL). Because drugs are tethered to high concentration on the surface of cells, a longstanding question has been whether the tethered drug can retain its molecular specificity for the receptor of interest. Additionally, given that cells can be closely apposed to one another, it has been uncertain whether a tethered drug can maintain cellular specificity, without reaching across to act on receptors on adjacent cells. In this study, we investigated the effects of our new YM90K.1$^{DART.2}$ reagent on excitatory synaptic function in genetically identified brain slices of the ventral tegmental area (VTA, Dopamine neurons) and the striatum (D1- and D2- type neurons), as well as on unitary synaptic function in defined pre- or post-synaptic cells of cultured hippocampal neurons. With regard to molecular specificity, our patch clamp recordings reveal that YM90K.1$^{DART.2}$ quickly blocks AMPA receptor-mediated excitatory post-synaptic currents (EPSCs), without influencing $GABA_A$ receptor-mediated inhibitory post-synaptic currents (IPSCs). To explore the potential impact of DART.2 to act trans-cellularly, we performed paired patch recordings in cultured hippocampal neurons expressing either the $^{+}$HTP or a 'double dead' (^{dd}HTP) construct. Post-synaptic AMPAR-mediated unitary excitatory postsynaptic potentials (uEPSPs) induced by a single action potential stimulation from pre-synaptic cell using paired recording methods. We find that YM90K.1$^{DART.2}$ successfully blocks uEPSPs only if the post-synaptic neuron expressed $^{+}$HTP. In contrast, expression of $^{+}$HTP on the presynaptic cell resulted in no impact of tethered drug. In summary, our newly developed YM90K.1$^{DART.2}$ reagent offers specific modulation of excitatory post-synaptic function without affecting pre-synaptic neurotransmission. This reagent paves the way for cellular and synaptic specificity in future studies on neuronal circuitry and animal behavior.

Disclosures: **H. Yan:** None. **B.C. Shields:** None. **S.S.X. Lim:** None. **S. Burwell:** None. **M. Tadross:** None.

Poster

PSTR134. Parkinson's Disease: Small Molecule Therapeutics

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR134.09/G9

Topic: C.03. Parkinson's Disease

Support: NIH-T32GM144895

Title: Novel NOX inhibitors mitigate Parkinson's Disease Related pathology in PFF-injected mice.

Authors: *D. WHEELER[1], K. OFORI[1], D. VERMA[1], A. GHOSH[1], S. MOON[2], Y.-H. KIM[1]; [1]Delaware State Univ. Dept. of Biol. Sci., Dover, DE; [2]AptaBio Theraputics, Heungdeok, Korea, Republic of

Abstract: Parkinson's disease (PD) is a progressive neurodegenerative motor disorder without an applicable therapeutic to prevent dopaminergic neuronal loss in the nigrostriatal pathway. An essential hallmark of PD pathology is the formation of mis-folded protein aggregates called Lewy bodies. Therefore, it has been prioritized to develop a therapeutic to halt the Lewy body formation and prevent overall dopaminergic neuronal loss. In the down-stream pathology, oxidative stress-mediated damage has been often reported as a critical mechanism to trigger neuronal loss in PD pathology. Therefore, the suppression of reactive oxygen species (ROS) generation can be an effective approach to prevent PD progression. Here, we are assessing the efficacy of novel nicotinamide adenine phosphate (NADPH) oxidase (NOX) inhibitors, the compound-11 & 19 (C-11 & 19) in N27 rat dopaminergic cells and PD mouse models. *In vitro* assessment showed that both compounds successfully reduced cytotoxicity and simultaneously enhanced cell viability against the exposure to α-synuclein preformed fibrils (PFF) induced protein aggregation at various concentrations (1 nM-10 μM). The levels of PFF-induced ROS and protein aggregation as shown in the ROS and Thioflavin-T assays were also substantially reduced at the optimal concentration of NOX inhibitors (around 100 nM) compared with PFF only. The C-11 or 19 was gavaged at two different doses (5 or 25 mg/kg) to stereotaxically PFF-injected mice for 6 or 10 weeks, respectively, and behavioral tests were assessed for the last two weeks before the sacrifice. Before treatment, the PFF-injected mice presented obvious motor deficits, loss in neuromuscular coordination, and functional complications when compared to their wild-type counterparts. Consistent oral treatment demonstrated that both C-11 and -19 successfully alleviated motor deficits in all behavioral analyses, such as hindlimb clasping, pole test, rotarod, nesting, and grooming. In the following immunohistochemical analyses, NOX inhibition was effectively prevented or reversed the neuronal loss in the nigrostriatal pathway. These results suggest that the inhibition of NOX-1, 2, and 4 is a viable potential therapeutic target for preventing or at least partially reversing PD pathology.

Disclosures: **D. Wheeler:** None. **K. Ofori:** None. **D. Verma:** None. **A. Ghosh:** None. **S. Moon:** None. **Y. Kim:** None.

Poster

PSTR134. Parkinson's Disease: Small Molecule Therapeutics

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR134.10/G10

Topic: C.03. Parkinson's Disease

Title: New small molecule inhibitors of α-synuclein aggregation and toxicity

Authors: *S. ARYA[1], V. MATHUR[1], M. KOKES[1], E. SHAO[1], J. SCHWARZ[2], K. BURK[3], L. WHITEHEAD[3], A. SINGH[5,4], K. PLANEY[4];
[1]Preclinical Team, [2]Chem., [3]Computat. Discovery, [4]Acelot, Palo Alto, CA; [5]Dept. of Computer Science, Biomolecular Sci. and Engin., Univ. of California Santa Barbara, Santa Barbara, CA

Abstract: Background: Aberrant aggregation and accumulation of α-synuclein in the brain is the pathological hallmark of a group of neurodegenerative disorders known as synucleinopathies that include Parkinson's disease (PD), multiple system atrophy (MSA), and dementia with Lewy bodies (DLB). Therefore, aggregates of α-synuclein are a potential therapeutic target for treating synucleinopathies. Although a few drug candidates are in the development pipeline, there is currently no approved treatment that could halt or slow disease progression. Furthermore, no clinical stage drug has been shown to directly disrupt α-synuclein aggregates.**Objective**: To identify small molecule inhibitors of α-synuclein aggregation and to show their *in vitro* potency and mechanism of action.**Methods:** Using our novel joint pharmacophore space (JPS) machine learning platform, we identified a subset of hit compounds that specifically bind to amyloid oligomers of α-synuclein and other neurodegenerative disease relevant proteins such as Aβ42, Tau and TDP43. We evaluated 150 compounds derived from the JPS platform in a robust, high-throughput fluorescence-based, cell-free primary screen for disruption of α-synuclein amyloid aggregation. We further evaluated the interactions of select compounds with α-synuclein aggregates by monitoring aggregate size changes using dynamic light scattering (DLS). The hit compounds from cell-free assays were finally tested in cell-based a-synuclein oligomer toxicity assay.**Results:** We have identified a novel small molecule ACE339 and its structural analogs as lead compounds that effectively disassemble pre-formed α-synuclein amyloid aggregates. Furthermore, in our cell-based assay, we show that ACE339 and its analogues can rescue α-synuclein oligomer dependent neuronal toxicity. *In vitro* ADME (absorption, distribution, metabolism, and excretion) and mouse PK studies show high microsomal stability, high gut permeability and excellent brain penetrance for ACE339. We are currently testing the potency of our lead compounds, the mechanism of action and their off-target activity.**Conclusion:** ACE339 and its structural analogs, disaggregate recombinant alpha-synuclein aggregates *in vitro*, and mitigate alpha-synuclein cytotoxicity in neuronal cells. These compounds have favorable pharmacokinetic properties and brain permeability. Thus, these compounds can be further characterized and developed as potential therapeutics for synucleinopathies.

Disclosures: **S. Arya:** A. Employment/Salary (full or part-time); Acelot. **V. Mathur:** A. Employment/Salary (full or part-time); Acelot. **M. Kokes:** None. **E. Shao:** None. **J. Schwarz:** None. **K. Burk:** None. **L. Whitehead:** None. **A. Singh:** None. **K. Planey:** None.

Poster

PSTR134. Parkinson's Disease: Small Molecule Therapeutics

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR134.11/H1

Topic: C.03. Parkinson's Disease

Support: MJFF

Title: Ezeprogind (azp2006), a new progranulin/prosaposin modulator, as an innovative therapeutic treatment for parkinson's disease.

Authors: *P. VERWAERDE[1], C. ESTRELLA[2], N. CALLIZOT[2], A. HENRIQUES[3]; [1]Alzprotect SAS, Loos, France; [2]Alzprotect, Loos, France; [3]Pharmacol., Neuro-Sys, GARDANNE, France

Abstract: Parkinson's disease (PD) is the second most common neurodegenerative disorder of dopaminergic neurons in the substantia nigra pars compacta, causing a loss of dopamine (DA) in the striatum and the presence of intraneuronal inclusions, named Lewy bodies. While DA replacement therapies can reduce motor symptoms, current therapies do not modify the disease progression. Around 5-15% of PD patients have GBA mutations (lysosomal glucocerebrosidase enzyme, GCase), becoming the most important genetic risk factor for PD (more frequently than other genes like LRRK2, SNCA and PARK2). Progranulin (PGRN), is a secreted growth factor widely expressed throughout the human body, notably in nervous system. PGRN regulates neuroinflammation, neurite branching, outgrowth and it's necessary for maintaining lysosomal function, facilitating the activity of several lysosomal enzymes, including the critical protease cathepsin D. PGRN lysosomal availability depends on its interaction with prosaposin (PSAP). PSAP is essential not only for PGRN transport but also for GCase activation. AZP2006 is a small molecule displaying neuroprotective properties currently under clinical development for Progressive Supranuclear Palsy. Its effect was shown to involve PGRN/PSAP complex, binding and stabilizing it and then increasing the central PGRN levels. Although there is some incertitude about its precise mode of action, it has been shown that under stress conditions, AZP2006 targeted the lysosomes and was able to help lysosomal function and homeostasis. In addition, in absence of PGRN or PSAP, AZP2006 neuroprotective effects were fully abolished. In this study, we investigated AZP2006 effects in *in vitro* and *in vivo* PD models. Using primary cultures of mesencephalic neurons from rat embryos injured by different mitochondrial (MPP^+) or lysosomal toxins (CBE an irreversible inhibitor of GCase), or by α-syn pre-formed fibrils (PFF), AZP2006 was able to rescue TH neurons, prevent lysosomal burden and the α-syn aggregation. These effects were driven by PGRN/PSAP leading to the restoration of lysosomal function. *In vivo*, in aged animals using moderate α-syn PFF administration in the SNpc, combined or not

with a slight lysosomal GCase deficiency induced by CBE, AZP2006 was able to prevent the lesion after a 4-week oral daily treatment. Motor dysfunctions were restored after treatment at a same extend of Ambroxol (used as positive control). The lysosomal stress was abolished and the inflammation was decreased. PGRN brain levels were increased after the treatment. Altogether, these results make of AZP2006 a serious candidate for PD treatment and synucleopathies.

Disclosures: **P. Verwaerde:** A. Employment/Salary (full or part-time); Alzprotect/full. **C. Estrella:** A. Employment/Salary (full or part-time); Alzprotect/full. **N. Callizot:** A. Employment/Salary (full or part-time); Alzprotect/full. **A. Henriques:** A. Employment/Salary (full or part-time); neuro-sys/full.

Poster

PSTR134. Parkinson's Disease: Small Molecule Therapeutics

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR134.12/H2

Topic: C.03. Parkinson's Disease

Support: The Penn State Translational Brain Research Center
Children's Miracle Network Research Grant (2022–2023)
NIH RF1 AG071675

Title: Dynamic nigral iron changes after chronic levodopa treatment in a rat Parkinson's model

Authors: G. DU, M. ZHOU, L. BRANSOM, X. HUANG, R. B. MAILMAN, ***Y. YANG**; Penn State Univ. Col. of Med., Hershey, PA

Abstract: Parkinson's disease (PD) is marked pathologically by loss of dopamine neurons in the substantia nigra pars compacta. Despite effective dopamine replacement therapy, patient symptoms and dopamine neuronal loss keep progressing, eventually leading to patients being wheelchair bound, often developing dementia. Iron is an essential trace metal, but both post-mortem and modern in vivo MRI imaging approaches have reported increased iron in the substantia nigra of PD patients. The increases in nigral iron content have been postulated to be a causative mechanism in PD because iron is known to catalyze oxidative events that can lead to cellular damage. Current study therefore aimed to examine if levodopa, the gold standard symptomatic pharmacological treatment of PD, would change the course of nigral iron accumulation. The results would help clarify a decades old controversy: if levodopa can provide neuron protective effects via slowing down the nigral iron accumulation or actually have potential toxicity by causing more nigral iron accumulation. Hemiparkinsonian rats were created by unilateral injection of 6-hydroxydopamine into the medial forebrain bundle. Levodopa was subcutaneously administered at 100 mg/kg per day for 15 days. Nigral iron was measured before and after the chronic levodopa treatment through in vivo MRI imaging, using apparent relaxation rate (R2*) images. The result showed that the nigral iron was significantly increased after the levodopa treatment, especially in the brain that had mild dopamine neuronal loss. This result

suggested that levodopa, while essential for patient welfare and for positive effects in normalizing brain circuitry, may concomitantly start a cascade that increases neurodegeneration evidenced by iron accumulation. Importantly, this potential toxicity may be prominent at the early stage PD patients. The current pilot study provided a foundation for a potentially paradigm-shifting hypothesis: nigral iron levels that either reflect or catalyzing toxicity increase is a consequence of PD treatment levodopa. The corollary is to support the future clinical hypothesis that a drug, that could equal levodopa in symptomatic efficacy but does not contain reactive structural moieties, would not accelerate degeneration of dopamine neurons. In the absence of the long-sought cure or disease-arresting therapy, this could be a lead to the greatest advance for PD since levodopa became available a half-century ago.

Disclosures: G. Du: None. **M. Zhou:** None. **L. Bransom:** None. **X. Huang:** None. **R.B. Mailman:** None. **Y. Yang:** None.

Poster

PSTR134. Parkinson's Disease: Small Molecule Therapeutics

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR134.13/H3

Topic: C.03. Parkinson’s Disease

Support: TL1TR002016

Title: Contribution of levodopa to brain iron accumulation

Authors: *R. O. SERPA[1], K. PALSA[2], J. R. CONNOR[2];
[1]Penn State Col. of Med. Neurosci. Grad. Program, Hershey, PA; [2]Neurosurg., Penn State Col. of Med., Hershey, PA

Abstract: The antiparkinson drug levodopa (**L-DOPA**) is the current standard of treatment for Parkinson’s Disease (**PD)** and can be used as either a monotherapy or in combination with monoamine oxidase-B (**MAO-B**) inhibitors, such as selegiline to ameliorate dopamine depletion. Despite L-DOPA’s recognition as the gold standard for PD management, our group has recently made a fundamental observation of iron accumulation in the substantia nigra (**SN**) of PD patients following L-DOPA but not selegiline administration. These findings indicate that the type of antiparkinson drug may be correlated with nigral iron increase. Evidence suggests that nigral iron accumulation is increased in PD patients, decreasing dopamine thus exacerbating the progression of neurodegeneration. Though, it remains unclear whether increased iron in the SN is from PD itself or from L-DOPA treatment. Our current study tests the hypothesis that L-DOPA exposure will cause an increase in iron transport from the blood-brain barrier (**BBB**) to the brain in a BBB model. The effects of L-DOPA on iron-bound transferrin (**^{55}Fe-Tf**), a major iron source for the brain were investigated using endothelial cells (**ECs**) derived from human-induced pluripotent stem cells (**hiPSC**). ECs cultured in Transwell inserts were treated with 100μM L-DOPA, 100μM L-DOPA+0.01μM selegiline, or 0.01μM selegiline. ^{55}Fe-Tf transport and uptake from

the ECs were quantified via liquid scintillation counter. Our results showed a significant increase of ^{55}Fe-Tf transport in L-DOPA versus control at hour 2 (*p<0.05) supporting the hypothesis. In addition, we found less ^{55}Fe-Tf content in the ECs in both the 100μM L-DOPA (**p<0.01) and 100μM L-DOPA+0.01μM selegiline (***p<0.001) when compared to the control and no differences were observed in the selegiline group after 24 hours. The data suggests that L-DOPA affects iron transport by modifying the iron status of the ECs and mediating iron transport. The results suggest that the common antiparkinson treatment medication, L-DOPA, alters iron homeostasis that may promote neurodegeneration through iron accumulation; a novel concept requiring further study.

Disclosures: R.O. Serpa: None. **K. Palsa:** None. **J.R. Connor:** None.

Poster

PSTR135. Muscle Diseases and Hereditary Spastic Paraparesis

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR135.01/H4

Topic: C.06. Neuromuscular Diseases

Title: Improved kinematic gait phenotype in hDMDdel52/mdx mouse model of Duchenne muscular dystrophy after treatment with antisense oligonucleotide BMN351

Authors: J. PUOLIVÄLI[1], T. BRAGGE[1], Y. QI[2], J. HENSHAW[2], K. LARIMORE[2], C. CARTER[3], P. FANT[4], ***J. OKSMAN**[1], L. WETZEL[2], M. SIGG[2], C. A. O'NEILL[2], T. OPPENEER[2];
[1]Charles River Discovery Services, Kuopio, Finland; [2]BioMarin Pharmaceut. Inc., San Rafael, CA; [3]Charles River Labs., Mattawan, MI; [4]Charles River Labs. France Safety Assessment, St. Germain-Nuelles, France

Abstract: Duchenne muscular dystrophy (DMD) is a chromosome X-linked progressive muscle disease caused by lack of functional dystrophin. BMN 351 is a next-generation antisense oligonucleotide developed for the treatment of DMD. BMN 351 induces skipping on exon 51 leading to production of near-full-length dystrophin protein. Here we studied the effects of BMN 351 in a hDMDdel52/*mdx* mouse model of DMD that lacks both mouse and human dystrophin, which leads to a dystrophic phenotype.
hDMDdel52/*mdx* mice were dosed via intravenous injection (IV) with vehicle or BMN 351 for 13 or 25 weeks. Vehicle treated C57BL/6J mice were used as controls. Kinematic gait analysis was conducted at various time points after dosing. Movement of the animal was captured from 3 dimensions using a high-speed camera. The analyzed parameters included general gait, body posture and balance, and fine motor skills. At the terminal endpoint, blood samples were analyzed for clinical chemistry and hematology, and tissue samples were analyzed for histopathology, percent exon skipping, and dystrophin levels.
Vehicle treated hDMDdel52/*mdx* mice showed increased levels of blood clinical pathology parameter such as ALT, AST, CK, and LDH, histopathological muscle injuries, and deficits in

the kinematic gait analysis compared to age-matched control mice, consistent with the previously observed phenotype. Treatment with BMN 351 resulted in increased levels of exon skipping and dystrophin levels in hDMDdel52/mdx mice. Increased dystrophin levels were associated with normalized blood clinical pathology parameters and a dose- and time-dependent reduction in model-related skeletal and myocardial muscle pathology. Importantly, the behavioral phenotype of hDMDdel52/mdx mice, as studied by kinematic gait analysis, was reversed by BMN 351 treatment.
These results show that treatment with BMN 351 significantly improves the biochemical, pathological, and behavioral phenotype of hDMDdel52/mdx mice and supports moving into clinical studies in patients with DMD.

Disclosures: **J. Puoliväli:** A. Employment/Salary (full or part-time); Charles River Discovery Services. **T. Bragge:** A. Employment/Salary (full or part-time); Charles River Discovery Services. **Y. Qi:** A. Employment/Salary (full or part-time); BioMarin Pharmaceutical Inc. **J. Henshaw:** A. Employment/Salary (full or part-time); BioMarin Pharmaceutical Inc. **K. Larimore:** A. Employment/Salary (full or part-time); BioMarin Pharmaceutical Inc. **C. Carter:** A. Employment/Salary (full or part-time); Charles River Laboratories. **P. Fant:** A. Employment/Salary (full or part-time); Charles River Laboratories France Safety Assessment. **J. Oksman:** A. Employment/Salary (full or part-time); Charles River Discovery Services. **L. Wetzel:** A. Employment/Salary (full or part-time); BioMarin Pharmaceutical Inc. **M. Sigg:** A. Employment/Salary (full or part-time); BioMarin Pharmaceutical Inc. **C.A. O'Neill:** A. Employment/Salary (full or part-time); BioMarin Pharmaceutical Inc. **T. Oppeneer:** A. Employment/Salary (full or part-time); BioMarin Pharmaceutical Inc.

Poster

PSTR135. Muscle Diseases and Hereditary Spastic Paraparesis

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR135.02/H5

Topic: C.06. Neuromuscular Diseases

Support: Canada Research Chair Tier II to VAF
Brock-Niagara Validation, Prototyping and Manufacturing Institute Award in partnership with AMO Pharma to VAF

Title: Tideglusib mitigates Alzheimer's disease-like pathology in aged D2 mdx mice.

Authors: *E. N. COPELAND[1], A. MOHAMMAD[2], B. M. MARCELLA[1], R. W. BARANOWSKI[1], R. E. K. MACPHERSON[2], V. A. FAJARDO[1];
[1]Kinesiology, [2]Hlth. Sci., Brock Univ., St Catharines, ON, Canada

Abstract: Introduction: Duchenne muscular dystrophy (DMD) is an X-linked, progressive muscle wasting disorder, occurring in approximately 1 in 5000 boys. In addition to muscular deficits, one third of patients also experience cognitive challenges such as memory loss.

Recently, the preclinical model of DMD, the DBA/2J (D2) *mdx* mouse, has been found to have an Alzheimer's disease (AD)-like pathology with reduced recognition memory and increased biochemical markers of amyloid beta (Aβ) production. The mechanisms behind this pathology are unknown; however, glycogen synthase kinase 3β (GSK3β) is a well-known contributor to AD and has also been implicated in DMD muscle pathology. Tideglusib is a non-ATP competitive GSK3 inhibitor, currently in clinical trials for AD and myotonic dystrophy type 1 (DM1). Recent work from our group has shown tideglusib treatment to improve muscle health in D2 *mdx* mice and thus its ability to improve brain health is also of interest. Here, we aimed to determine whether short-term tideglusib treatment can improve AD-like pathology in D2 *mdx* mice via GSK3β inhibition.
Methods: 9–10-week-old D2 WT and *mdx* mice were purchased from Jackson Laboratories and aged to 26 weeks. Mice were separated into the following groups: 1) WT healthy controls, 2) *mdx* vehicle, and 3) *mdx* tideglusib (10 mg/kg/day via oral gavage). Tideglusib treatment lasted a total of 4 weeks. Prior to euthanasia, a novel object recognition test (NORT) was performed to assess spatial memory. Hippocampus and serum samples were collected from each mouse for *in vivo* studies, including BACE1 activity assays, Aβ ELISAs.
Results: Tideglusib improved cognitive function in aged *mdx* mice with significantly higher exploration time of novel objects in the NORT. However, BACE1 activity was not different between vehicle and treatment groups. Additionally, we measured serum levels of Aβ as a marker of its vascular clearance. Our results show that tideglusib treated *mdx* mice had greater levels of circulating Aβ compared with vehicle treated mice.
Conclusion: Tideglusib treatment improved cognitive function in aged D2 *mdx* mice and this was associated with an increase in circulating Aβ. Future studies will investigate Aβ clearance and other cellular mechanisms that may influence these cognitive alterations.

Disclosures: E.N. Copeland: C. Other Research Support (receipt of drugs, supplies, equipment or other in-kind support); This project was funded partly by AMO Pharma, who distributes tideglusib and provided the tideglusib for this project. **A. Mohammad:** C. Other Research Support (receipt of drugs, supplies, equipment or other in-kind support); This project was funded partly by AMO Pharma, who distributes tideglusib and provided the tideglusib for this project. **B.M. Marcella:** C. Other Research Support (receipt of drugs, supplies, equipment or other in-kind support); This project was funded partly by AMO Pharma, who distributes tideglusib and provided the tideglusib for this project. **R.W. Baranowski:** C. Other Research Support (receipt of drugs, supplies, equipment or other in-kind support); This project was funded partly by AMO Pharma, who distributes tideglusib and provided the tideglusib for this project. **R.E.K. MacPherson:** C. Other Research Support (receipt of drugs, supplies, equipment or other in-kind support); This project was funded partly by AMO Pharma, who distributes tideglusib and provided the tideglusib for this project. **V.A. Fajardo:** B. Contracted Research/Research Grant (principal investigator for a drug study, collaborator or consultant and pending and current grants). If you are a PI for a drug study, report that research relationship even if those funds come to an institution.; AMO Pharma. C. Other Research Support (receipt of drugs, supplies, equipment or other in-kind support); This project was funded partly by AMO Pharma, who distributes tideglusib and provided the tideglusib for this project.

Poster

PSTR135. Muscle Diseases and Hereditary Spastic Paraparesis

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR135.03/H6

Topic: C.06. Neuromuscular Diseases

Support: Grant-in-Aid for Scientific Research 19H04064, 16H03266, 243900228, 21200023, 21591102
AMED Leap JP21mg00100007
Takeda Science Foundation

Title: Proteolytic ectodomain shedding of muscle-specific tyrosine kinase in myasthenia gravis

Authors: *K. SHIGEMOTO, S. MORI, T. OMURA, H. TSUMOTO, Y. MIURA;
Tokyo Metro Inst., Tokyo, Japan

Abstract: Autoantibodies to muscle-specific tyrosine kinase (MuSK) proteins at the neuromuscular junction (NMJ) cause refractory generalized myasthenia gravis (MG) with dyspnea more frequently than other MG subtypes. However, the mechanisms via which MuSK, a membrane protein locally expressed on the NMJ of skeletal muscle, is supplied to the immune system as an autoantigen remains unknown. Here, we identified MuSK in both mouse and human serum, and the amount was dramatically increased in mice with motor nerve denervation and in MG model mice. Peptide analysis by liquid chromatography-tandem-mass spectrometry (LC-MS/MS) confirmed the presence of MuSK in both human and mouse serum. Furthermore, patients with MG have significantly higher amounts of MuSK in serum than healthy controls. Our results indicated that secretion of MuSK proteins from muscles into the bloodstream was induced by ectodomain shedding triggered by neuromuscular junction failure. The results may explain why MuSK-MG is refractory and causes rapid muscle atrophy in some patients due to the denervation associated with Ab-induced disruption of neuromuscular transmission at the NMJ. Such discoveries pave the way for new MG treatments, and MuSK may be used as a biomarker for other neuromuscular diseases in preclinical studies, clinical diagnostics, therapeutics, and drug discovery.

Disclosures: K. Shigemoto: None. **S. Mori:** None. **T. Omura:** None. **H. Tsumoto:** None. **Y. Miura:** None.

Poster

PSTR135. Muscle Diseases and Hereditary Spastic Paraparesis

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR135.04/H7

Topic: C.06. Neuromuscular Diseases

Title: Age-related differences in electromyographic coherence of rotator cuff and deltoid muscles during fatigue

Authors: *H. ZHU, X. YU, L. GRIFFIN;
Univ. of Texas, Austin, Austin, TX

Abstract: Rotator cuff muscles play an essential role in stabilizing the humeral head during contraction of the deltoid muscles. Many older adults suffer from limited shoulder mobility, which may be caused by imbalances in activation across the rotator cuff muscles and/or with the deltoid muscles. Neuromuscular fatigue can further alter muscle force imbalances, increasing the risk of developing rotator cuff disease. EMG coherence analysis is a measure of common fluctuations in neural drive to motor neuron pools of muscle pairs. It is possible that patterns of central oscillations of neural drive may change with age. The aim of this study was to compare EMG-EMG coherence across rotator cuff and deltoid muscles during fatigue in young and olde adults. 15 asymptomatic young (25.1 ± 9.9 yrs, 7 males) and 11 asymptomatic old (70.1 ± 10.0 yrs, 4 males) adults participated. Surface EMG was recorded from the infraspinatus (IS) and middle deltoid (MD) muscles. Intramuscular EMG was recorded from the supraspinatus (SS) muscle. Participants performed an isometric fatiguing contraction at 30° scaption at 25% maximal voluntary contraction until endurance limit. The first 30s and last 30 s were defined as the was defined as the pre-fatigue and fatigue phases. Z-transformed pooled coherence of each muscle pair (SS-IS, SS-MD, and IS-MD) in the delta (2-5 Hz) and beta (15-35 Hz) bands in two phases were compared with two-way repeated measures ANOVAs (Group x Time) with Bonferroni post-hoc analysis. In young group, increased EMG RMS amplitude and decreased slopes of EMG median frequency were found for all three muscles with fatigue. Whereas, in old group, the slope of EMG median frequency only declined in the IS muscle. For the SS-IS pair, a main effect of Group was found in the beta band ($p<0.01$) between young and old adults. Both groups increased coherence with fatigue in both the delta and beta bands; however, the old group had lower coherence in both bands. For the SS-MD pair, the young group increased coherence with fatigue in both the delta band and beta band ($p<0.001$); whereas coherence decreased in both the delta ($p<0.05$) and beta ($p<0.05$) bands with fatigue in the older group. No significant differences with fatigue were found in the IS-MD pair in either group. The increase in coherence of SS-IS pair in the delta band with fatigue indicates greater common drive within rotator cuff muscles in both two groups. While in the beta band, the reduction in coherence of SS-MD pair during neuromuscular fatigue indicates diminished shared neural inputs to the rotator cuff and deltoid muscles in the older group. Thus, this may be a contributing factor to the development of altered shoulder mobility and pain in older adults.

Disclosures: H. Zhu: None. **X. Yu:** None. **L. Griffin:** None.

Poster

PSTR135. Muscle Diseases and Hereditary Spastic Paraparesis

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR135.05/H8

Topic: C.06. Neuromuscular Diseases

Support: Miur, grant RM12117A613F246C

Title: Sensorimotor peripheral innervation in Duchenne Muscular Dystrophy: spotlight on the sciatic nerve of dystrophic mdx mice

Authors: *M. DE STEFANO[1], S. DI NUZZO[2], F. MASTROSTEFANO[2], M. SOLIGO[3], V. FERRETTI[2], V. MAGNAGHI[4];
[1]Biol. and Biotechnologies, [2]Biol. and Biotechnologies "Charles Darwin", Sapienza Univ. of Rome, Roma, Italy; [3]Inst. of Translational Pharmacol., Natl. Res. Council of Italy (CNR), Rome, Italy; [4]Dept. of Pharmacol. and Biomolecular Sci., Univ. of Milan, Milan, Italy

Abstract: Duchenne muscular dystrophy (DMD) is an X-linked neuromuscular disease, characterized by the lack of dystrophin (Dp427), a cytoskeletal protein expressed in skeletal muscles and several other cell types. Along or in substitution of Dp427, different tissues other than muscles may also express shorter dystrophin isoforms, as in the peripheral nervous system (PNS), where myelinating Schwann cells (SCs) contain the Dp116 isoform. By binding to a large multiprotein complex (DGC), described as similar in muscle and organized around the α- and β-dystroglycan (DG) dimer, Dp116 contributes to myelin integrity and nerve fiber conduction. The structural and functional integrity of nerve fibers relies on axon-SC intense cross-talk, a well-known mediator of which is the neurotransmitter GABA. To unveil the effects that DMD has on peripheral sensory-motor nerves, we analyzed in the sciatic nerve of *mdx* mice, a model of DMD, expression and localization of myelin proteins (e.g. MBP, P0, PMP22, MAG), Dp116, proteins of the DGC (dystrobrevin, beta-DG), as well as inotropic GABAA (α4, β3, δ) and metabotropic GABAB-1b receptor subunits. Compared to wild-type, in *mdx* mice, the level, distribution and intensity of immunolabeling of all proteins analyzed, except for Dp116, were significantly reduced, indicating alteration of myelin integrity and SCs-axons crosstalk. This was also confirmed by parallel electrophysiological studies showing a significant reduction in Abeta-mechanoreceptor fiber excitability. Furthermore, gel zymography and Western immunoblot demonstrated activation of the pro-inflammatory extracellular matrix metalloproteinase (MMP) 2, which targets both alpha and beta-DG, suggesting that cleavage of DG within the myelin sheet, induced by retrograde inflammation from muscle to nerve, could be an upstream determinant for Dp116-DGC degradation and consequent deregulation of SC-axon signaling. Altogether, these findings support the hypothesis of significant alterations of the neuron-SC cross-talk in peripheral nerves of dystrophic subjects, an aspect deserving consideration in DMD therapeutic strategies.

Disclosures: M. De Stefano: None. **S. Di Nuzzo:** None. **F. Mastrostefano:** None. **M. Soligo:** None. **V. Ferretti:** None. **V. Magnaghi:** None.

Poster

PSTR135. Muscle Diseases and Hereditary Spastic Paraparesis

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR135.06/H9

Topic: C.06. Neuromuscular Diseases

Support: Carter Foundation for Neurological Research
National Institutes of Health
Massachusetts General Hospital

Title: Modeling axonal degeneration in spastic paraplegias type 3A with human pluripotent stem cells

Authors: *G. THAKUR;
Biomed. Sci., Univ. of Illinios, Chicago, Rockford, IL

Abstract: Hereditary spastic paraplegias (HSPs) are a heterogeneous group of neurological disorders caused by a length-dependent axonopathy of long corticospinal projection neurons. SPG3A, the most prevalent early-onset form of HSP, is caused by the mutations in the *ATL1* gene that encodes the Atlastin-1 protein. By transducing patient fibroblast cells with pluripotent factors, we successfully generated induced pluripotent stem cells (iPSCs) from two SPG3A patients with mutations at different regions (p.P342S and p.M408T). After characterizing the iPSC lines, we differentiated these patient stem cells into cortical projection neurons, the cell types prominently affected in the patients. Patient iPSC-derived cortical neurons exhibited reduced axonal outgrowth and accumulated axonal swellings, recapitulating disease-relevant axonal defects. To monitor axonal degeneration in long-term cultures, we examined the levels of phosphorylated neurofilament heavy chain (pNFH) in culture medium using a high sensitive pNFH ELISA kit. Increased release of pNFH has been reported in common neurodegenerative diseases and serves as a potential biomarker to monitor axonal degeneration. Interestingly, we observed a significant increase of pNFH in medium of SPG3A cortical neurons compared to that in control neuron medium, implying its potential as a marker to monitor axonal defects in SPG3A neurons. Finally, we examined the protective effects of LXR agonists on SPG3A patient iPSC-derived cortical projection neurons. Treatment with LXR agonists significantly mitigated the accumulated axonal swellings in long-term cortical neurons derived from SPG3A iPSCs as compared to control cortical neurons. The protective effects of the LXR agonist were further supported by the mitigation of increased pNFH levels and the rescue of subsequent apoptosis in SPG3A cortical PNs after LXR agonist treatment. Taken together, our data reveal the recapitulation of disease-relevant axonal defects in SPG3A patient iPSC-derived neurons and highlight the potential of LXR agonists in rescuing axonal and neuronal degeneration in SPG3A. The detailed mechanisms underlying axonal degeneration and the efficacy of LXR agonists *in vivo* in HSP animal models will be further examined in the future.

Disclosures: G. Thakur: None.

Poster

PSTR135. Muscle Diseases and Hereditary Spastic Paraparesis

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR135.07/H10

Topic: C.06. Neuromuscular Diseases

Support: The University of Queensland Amplify Fellowship
Australian Government Research Training Program Scholarships (RTP)
Westpac Scholars Trust
Commonwealth Tuition Fee Ofset Scholarships
Academy of Finland Grant 335527
Rebecca L Cooper Foundation

Title: Using super-resolution microscopy to decipher membrane trafficking in live neurons

Authors: *N. YAK[1,2], S. ABD ELKADER[1,2], G. BALISTRERI[1,3], M. JOENSUU[1,2];
[1]The Univ. of Queensland, Queensland Brain Inst., Brisbane, Australia; [2]The Univ. of Queensland, Australian Insitute for Bioengineering and Nanotechnology, Brisbane, Australia; [3]Fac. of Medicine, Medicum Res. Program, Univ. of Helsinki, Helsinki, Finland

Abstract: Our understanding of the detailed molecular mechanisms controlling membrane trafficking at the secretory pathway and in the synapse of murine primary neurons is incomplete due to the diffraction limit of light microscopy. To address this, our lab implements several super-resolution microscopy techniques such as Universal Point Accumulation in the Nanoscale Topography (uPAINT) (Giannone et al, *Methods Mol Biol. (*2013); Joensuu et al, *J Cell Biol.* (2016); Small et al, *Methods Mol Biol.* (2021)), subdiffractional Tracking of Internalized Molecules (sdTIM) (Joensuu et al, *J Cell Biol.* (2016); Small et al, *Methods Mol Biol.* (2021)), and single-particle-tracking Photo-Activated Localization Microscopy (sptPALM) (Manley et al, *Nat Methods.* (2013); Small et al, *Methods Mol Biol.* (2021)). sdTIM and uPAINT allow us to image and track single molecules of anti-GFP Atto647N-tagged nanobodies (At647N) bound to pHluorin-conjugated synaptic membrane proteins in live hippocampal neuron plasma membrane or following endocytosis into synaptic vesicles, respectively. In sptPALM, mEos2 fluorophores are stochastically activated, allowing imaging and tracking of single molecules inside live neurons. Using uPAINT, we aim to understand how autism spectrum disorder-associated mutations in actin binding protein alpha-Actinin-4 affect singe molecule mobility and nanoclustering of membrane receptors GluA1/2 in the postsynapse. My PhD project focuses on hereditary spastic paraplegia type 54 (HSP54). We aim to understand how loss of function mutations in lipid modifying enzyme DDHD2 affect synaptic membrane trafficking (using sdTIM), and secretory pathway membrane trafficking in-between compartments such as Golgi and Endoplasmic Reticulum Golgi Intermediate Compartment (ERGIC) (using sptPALM). Our results demonstrate that these techniques are powerful in deciphering the nanoscale changes in protein mobility in these disease conditions and provide us with detailed information about the cellular pathology of the conditions.

Disclosures: **N. Yak:** None. **S. Abd Elkader:** None. **G. Balistreri:** None. **M. Joensuu:** None.

Poster

PSTR135. Muscle Diseases and Hereditary Spastic Paraparesis

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR135.08/I1

Topic: C.06. Neuromuscular Diseases

Support: The University of Queensland Amplify Fellowship
Westpac Scholars Trust
Australian Government Research Training Program Scholarships (RTP)
Commonwealth Tuition Fee Ofset Scholarships
Academy of Finland Grant 335527

Title: Disturbed lipid metabolism in the neuropathology of hereditary spastic paraplegia type 54 (HSP54)

Authors: *S. ABD ELKADER[1,2], N. YAK[1,2], X. L. H. YONG[1], A. GAUDIN[1], J. HARMER[3], V. ANGGONO[1], G. BALISTRERI[1,4], M. JOENSUU[1,2];
[1]The Univ. of Queensland Queensland Brain Inst., Brisban, Australia; [2]Australian Inst. for Bioengineering and Nanotechnology, Brisbane, Australia; [3]Ctr. for Advanced Imaging, The Univ. of Queensland, Brisbane, Brisbane, Australia; [4]Fac. of Medicine, Medicum Res. Program, Univ. of Helsinki, , Finland, Helsinki, Finland

Abstract: The phospholipid and free fatty acid (FFA) composition in the neuronal membranes is vital for neuronal functions such as learning and memory acquisition (Joensuu et al., 2019 J Neurochem 153). However, the specific mechanisms by which neuronal activity influences the lipid landscape in the brain is not well understood, presenting an exciting area for further exploration. Interestingly, we have recently shown that the levels of saturated FFAs, in particular myristic acid (C14:0), increase significantly during neuronal stimulation *in vitro* in hippocampal neuronal cultures, and behavioral memory paradigms in mice *in vivo* (Akefe et al., 2023 bioRxiv preprint). This observation suggests a potential role for phospholipase A1 (PLA1) isoform DDHD2 activity in synaptic plasticity, hydrolyzing the release of saturated FFAs from neuronal membranes. Hereditary Spastic Paraplegia (HSP) type 54 is a neurodegenerative condition caused by mutations in the *DDHD2* gene in humans. The exact mechanisms by which *DDHD2* gene mutations cause HSP54 are not fully understood. Here, using super-resolution imaging, electron microscopy, Electron paramagnetic resonance (EPR), and cell biological approaches, we show that the genetic ablation of the *DDHD2* in mice (*DDHD2*$^{-/-}$) disturbs intracellular membrane trafficking in the secretory pathway and in the synapse of cultured hippocampal neurons. Similar effects can be observed after pharmacological inhibition of host protein N-myristoylation, indicating a direct role for disturbed protein lipidation in the neuropathology of HSP54.

Disclosures: S. Abd Elkader: None. **N. Yak:** None. **X.L.H. Yong:** None. **A. Gaudin:** None. **J. Harmer:** None. **V. Anggono:** None. **G. Balistreri:** None. **M. Joensuu:** None.

Poster

PSTR135. Muscle Diseases and Hereditary Spastic Paraparesis

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR135.09/I2

Topic: C.06. Neuromuscular Diseases

Support: The Myositis Association (90097118 to C.I.)
NIH (R01-NS095969 to P.C.W., R01-NS082563 to T.E.L., and R01-AR076390 to T.E.L. and P.C.W.)
Maryland Technology Development Corporation to P.C.W.
Muscular Dystrophy Association 630399 to T.E.L.
Johns Hopkins Discovery Award to T.E.L.
Peter and Carmen Lucia Buck Foundation to T.E.L. and P.C.W.

Title: Tdp-43 dependent cryptic exon derived neoepitopes as a novel diagnostic biomarker in muscle biopsies of inclusion body myositis patients

Authors: *C. IKENAGA, A. WILSON, A. P. MALLIKA, I. SINHA, G. BURNS, J. P. LING, A. M. CORSE, P. C. WONG, T. E. LLOYD;
Johns Hopkins Univ. Sch. of Med., Baltimore, MD

Abstract: Inclusion body myositis (IBM) is an idiopathic inflammatory myopathy with muscle pathology characterized by endomysial inflammation, rimmed vacuoles, and mislocalization of TDP-43. The loss of TDP-43-mediated splicing repression in myonuclei generates cryptic exons in IBM confirmed by both reverse transcription polymerase chain reaction and in situ hybridization. In most cases, cryptic exons are out-of-frame and degraded via nonsense-mediated decay. Utilizing a novel antibody recognizing cryptic peptide Hepatoma-Derived Growth Factor-Like protein 2 (HDGFL2), immunohistochemical analysis of muscle biopsies from 119 IBM patients revealed cryptic HDGFL2 in the myonuclei in 81 patients (68%). In contrast, cryptic HDGFL2 immunoreactivity was absent in 195 muscle biopsies from a variety of disease controls, except in one patient with Valosin-containing protein disease. Combining cryptic HDGFL2 immunoreactivity with the presence of rimmed vacuoles, the sensitivity for a diagnosis of IBM increased to 82% and specificity remained 99%. Notably, cryptic HDGFL2 transcripts and proteins were detected in both normal and abnormal appearing muscle fibers in IBM as judged by the hematoxylin and eosin stain, suggesting that the loss of TDP-43 splicing repression may occur in an early phase of the disease, preceding the appearance of rimmed vacuoles and protein aggregates in a myofiber. These results support the view that loss of TDP-43 occurs in skeletal muscle of IBM patients and suggest that immunostaining for the presence of the cryptic peptide HDGFL2 could be a potential early diagnostic marker for IBM patients.

Disclosures: C. Ikenaga: None. **A. Wilson:** None. **A.P. Mallika:** None. **I. Sinha:** None. **G. Burns:** None. **J.P. Ling:** None. **A.M. Corse:** None. **P.C. Wong:** None. **T.E. Lloyd:** None.

Poster

PSTR135. Muscle Diseases and Hereditary Spastic Paraparesis

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR135.10/I3

Topic: C.06. Neuromuscular Diseases

Support: NIH Grant AI093504
DoD Grant CB10721

Title: Reversal of neuromuscular weakness by modulating both central and peripheral motor synapses

Authors: B. T. MCCLINTIC[1], Z. D. CHANDLER[2], S. W. O'BRIEN[1], A. R. JACOBSON[1], ***P. M. MCNUTT**[2];
[1]Wake Forest Inst. for Regenerative Med., Wake Forest ISchool of Med., Winston-Salem, NC;
[2]Wake Forest Inst. for Regenerative Med., Wake Forest Sch. of Med., Winston Salem, NC

Abstract: Botulinum neurotoxin (BoNT) is a potent microbial toxin that causes muscle paralysis and death by asphyxiation. The only treatments for botulism are intubation and supportive care until respiratory function recovers, which can take weeks or longer. We have recently reported the FDA-approved drug 3,4-diaminopyridine (3,4-DAP) is the first small molecule antidote for clinical botulism. In rodent models of botulism, bolus administration of 3,4-DAP rapidly reverses toxic signs of botulism, while continuous infusion for two weeks had antidotal effects, without symptomatic rebound after stopping treatment. Although the therapeutic effects of 3,4-DAP are presumed to be caused by reversal of paralysis at diaphragm neuromuscular junctions (NMJ), the resulting effects on respiration are not understood. Here, we combined unrestrained whole-body plethysmography (UWBP) with arterial blood gas measurements to study the effects of 3,4-DAP and other aminopyridines on ventilation and respiration in the terminal stages of botulism in mice. Treatment of symptomatic mice with clinically relevant doses of 3,4-DAP restored ventilation in a dose-dependent manner, producing significant improvements in tidal volume, respiratory rate, and minute volume. Concomitant with improved ventilation, 3,4-DAP treatment reversed botulism-induced respiratory acidosis, restoring CO_2 and blood pH to normal physiological levels within 30 min. Having established that 3,4-DAP-mediated improvements in ventilation are correlated with improved respiration, we next used UWBP to quantitatively evaluate eight structurally related aminopyridines for therapeutic efficacy in BoNT-intoxicated mice. Multiple aminopyridines were identified with similar or improved therapeutic efficacies compared to 3,4-DAP. Notably, some aminopyridines preferentially enhanced tidal volume, while others preferentially enhanced respiratory rate. We interpret these results to indicate that aminopyridines can directly or indirectly modulate central respiratory circuits, as well as facilitate neurotransmission at the NMJ. In addition to contributing to a growing body of evidence supporting the use of aminopyridines in the treatment of clinical botulism, these findings are expected to inform the development of aminopyridine derivatives with improved pharmacological properties for the treatment of multiple neuromuscular indications.

Disclosures: B.T. McClintic: None. **Z.D. Chandler:** None. **S.W. O'Brien:** None. **A.R. Jacobson:** None. **P.M. McNutt:** None.

Poster

PSTR136. Neuroinflammation: HIV, HSV, COVID19, and Other Infections I

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR136.01/I4

Topic: C.07. Neurotoxicity, Inflammation, and Neuroprotection

Support: IDOR intramural grant
FAPERJ
FINEP
CNPq

Title: Protective properties of phytocannabinoids in neurocovid human model

Authors: *M. GUIMARAES[1,2], C. S. G. PEDROSA[2], H. A. BRAZ-DE-MELO[3], C. S. SANTOS[2], K. G. MAGALHÃES[3], S. K. REHEN[1,2];
[1]Inst. de Ciências Biomédicas, Univ. Federal do Rio de Janeiro, Rio de Janeiro, Brazil; [2]Inst. D'Or de Pesquisa e Ensino, Rio de Janeiro, Brazil; [3]Univ. de Brasilia, Brasilia, Brazil

Abstract: The prevalence of neuroinflammation in COVID-19 patients suggests that it may contribute to cognitive sequelae. Notably, one-third of COVID-19 survivors experience neurological sequelae, such as fatigue and cognitive deficits, the latter often referred to as "brain fog". This condition appears to be exacerbated in patients with metabolic syndrome. Our previous work demonstrated that SARS-CoV-2 can infect human neural tissue, leading to neuroinflammation. However, this infection is non-productive; it does not allow the release of new viral particles (Pedrosa et al. *Stem Cell Research* 54 (2021)). Here we have utilized 3D neuronal cellular preparations derived from human induced pluripotent stem cells to investigate the potential of cannabinoids, which possess anti-inflammatory and neuroprotective properties, as a possible treatment for NeuroCOVID. To test this hypothesis, we developed an approach for studying NeuroCOVID in the context of metabolic syndrome. This involved adding LPS and the Nucleocapsid protein (Ptn N) from SARS-CoV-2 to human neurospheroids. We observed significant neuroinflammation evidenced by the release of pro-inflammatory cytokines and a reduction in neuroplasticity following LPS + Ptn N treatment. Cytokines IL-1beta, TNF-alpha, and IL-6 increased from 2 to 72 hours of treatment (IL-1beta Control 58.94±6.37 pg/mL, Ptn N + LPS 130.2±7.62 pg/mL; TNF-alpha Control 106.1±20.14 pg/mL, Ptn N + LPS 1027±491.9 pg/mL; IL-6 Control 42.37±5.66 pg/mL, Ptn N + LPS 119.1±9.68 pg/mL) and there was a substantial decrease in the extension of neurites from plated neurospheres after 24 hours of LPS + Ptn N exposure (42.06±11.13% compared to control). Subsequently, we pre-treated or post-treated these brain spheroids with phytocannabinoids. We found that Delta9-tetrahydrocannabinol (THC) could reduce inflammation (IL-1beta 48.78±4.65 pg/mL; TNF-alpha 100.7±9.76 pg/mL; IL-6 33.37±6.92 pg/mL), while cannabidiol (CBD) and cannabigerol (CBG) could not. Furthermore, THC appeared to rescue the reduction in neurite extension after LPS + Ptn N treatment (100.2±22.09% compared to control). These findings hint at the possibility of considering phytocannabinoids as a treatment for cognitive deficits arising from NeuroCOVID.
Keywords:human induced pluripotent stem cells; cannabinoids; COVID-19

Disclosures: **M. Guimaraes:** None. **C.S.G. Pedrosa:** None. **H.A. Braz-de-Melo:** None. **C.S. Santos:** None. **K.G. Magalhães:** None. **S.K. Rehen:** None.

Poster

PSTR136. Neuroinflammation: HIV, HSV, COVID19, and Other Infections I

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR136.02/I5

Topic: C.07. Neurotoxicity, Inflammation, and Neuroprotection

Support: anonymous donor

Title: White matter changes observed in early Lyme disease are associated with fewer long-term symptoms

Authors: *C. MARVEL[1], P. A. NADKARNI[1], A. W. REBMAN[3], E. KOZERO[3], K. H. ALM[2], A. BAKKER[2], J. AUCOTT[3];
[1]Neurol., [2]Psychology, Johns Hopkins Univ., Baltimore, MD; [3]Med., Johns Hopkins Univ. Sch. of Med., Baltimore, MD

Abstract: BACKGROUND:Lyme disease is an inflammatory infection caused by *Borrelia burgdorferi* which is transmitted by the bite of an infected tick. After standard of care antibiotic therapy, 10-20% of patients will go on to develop a chronic syndrome of patient-reported symptoms, known as post-treatment Lyme disease syndrome (PTLD) that includes severe fatigue, arthritis, sleep disruption, neurocognitive impairments, and neurological symptoms. In a prior functional MRI study of patients with PTLD, we found white matter (WM) changes correlated with better outcomes. The nature of these WM changes, when they first occurred, and how they related to reduced symptoms, warranted further investigation.METHODS:Patients with acute Lyme disease (n=15) and healthy controls (HC, n=18) completed a working memory task in conjunction with a functional MRI scan within days of completing antibiotics. They received another fMRI scan 6 months later. Patients were categorized into those who “returned to health” (RTH) vs. those who developed symptoms of PTLD (sPTLD) based on their outcomes 6-12 months after infection and treatment. Each group’s fMRI data was compared to that of HC at the first (baseline) and second (6 months) MRI scans using $p < .005$. Regions of interest (ROIs) were created from between-group contrasts, using $p < .005$, $k = 10$ voxels. For Lyme patients, ROI values were correlated with the 36-Item Short Form Survey (SF-36) general health sub-score, which measured their self-reported global health status, using Pearson’s correlations, 2-tailed. RESULTS:The sPTLD group showed overall reduced neural activity relative to that of the HC group at baseline. At 6 months, their neural activity had increased and, therefore, had begun to normalize. By contrast, the RTH group at baseline showed extensive activity vs. HC, most of which was located in white matter. At 6 months, their neural activity had shifted to the frontal lobe and was represented primarily by gray matter. ROI values of the white matter regions at baseline positively correlated with their SF-36 scores at baseline and at 6 months later, $r(13) = .69$, $p = .004$ for both timepoints.IMPLICATIONS:White matter activations in Lyme disease

may reflect an appropriate healing response, and without it, patients face worse outcomes. This phenomenon may be relevant to other forms of infectious disease that share overlapping phenotypes with PTLD.

Disclosures: C. Marvel: None. **P.A. Nadkarni:** None. **A.W. Rebman:** None. **E. Kozero:** None. **K.H. Alm:** None. **A. Bakker:** None. **J. Aucott:** None.

Poster

PSTR136. Neuroinflammation: HIV, HSV, COVID19, and Other Infections I

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR136.03/I6

Topic: C.07. Neurotoxicity, Inflammation, and Neuroprotection

Support: NIH Grant EY09083
NIH Grant T32 AI 7519-24

Title: Neonatal herpes simplex virus infection leads to Alzheimer's-like cognitive decline and autophagy-mediated neurodegeneration in adult mice

Authors: *A. J. DUTTON[1,2], C. D. PATEL[2], S. A. TAYLOR[2], C. R. GARLAND[2], R. M. ALERS-VELAZQUEZ[2], J. MEHRBACH[2], D. A. LEIB[2], K. M. NAUTIYAL[3]; [2]Microbiology & Immunol., [1]Geisel Sch. of Med. at Dartmouth, Hanover, NH; [3]Psychological and Brain Sci., Dartmouth Col., Hanover, NH

Abstract: Alzheimer’s Disease (AD) is the fifth leading cause of death for Americans aged 65 and older. Despite the severity and prevalence of AD, however, the etiology of disease is still unknown. Previous work has identified female sex and apolipoprotein E ε4 (ApoE4) as leading risk factors for AD development, but these studies largely ignore environmental contributions to disease. More recently, microbial infection status has been implicated in the onset of cognitive decline and infection by herpes simplex virus type-1 (HSV-1) has been associated with AD development. To test the effect of HSV-1 infection on cognition in aging mice, we designed a low-dose neonatal HSV-1 (nHSV-1) infection model and tracked the impact of neonatally-contracted virus on the behavior of infected wild-type and human ApoE4-expressing mice. Brains of mice used for behavioral experimentation were assayed for markers of premature neurodegeneration and aging. Finally, to investigate the contribution of virus-mediated modulation of autophagy to the observed behavioral and neuropathologic phenotype, we assessed the long-term effects of infection by mutant HSV virus strains with deleted autophagy-regulatory domains. Wild-type mice neonatally infected with 100 plaque-forming units (pfu) of HSV-1 demonstrated measurable deficits in learning and memory tasks at four months of age compared to mock-infected controls. Specifically, nHSV-1 infected mice exhibited increased anxiety-like behavior on the Open Field Task (OFT), impaired novel object recognition on the Novel Object Recognition (NOR) task, and decreased hippocampal-specific memory performance on the Paired Associative Learning (PAL) paradigm. Furthermore, upon

examination of the hippocampus, infected mice showed increased deposition of misfolded protein, measured by Thioflavin-S staining. However, mice infected with herpes virus lacking the autophagy-regulating beclin-binding domain (BBD) in the γ34.5 gene did not exhibit learning and memory deficits or neuropathology at 4 months of age. Altogether, these findings suggest that the behavioral and neuropathological changes observed in our model may be mediated by low-dose viral infection and subsequent inhibition of host-cell autophagy. These results have implications for human health and AD research and are consistent with the idea that subclinical infections during the neonatal period may have long-term impact on neurodegenerative disease progression.

Disclosures: A.J. Dutton: None. **C.D. Patel:** None. **S.A. Taylor:** None. **C.R. Garland:** None. **R.M. Alers-Velazquez:** None. **J. Mehrbach:** None. **D.A. Leib:** None. **K.M. Nautiyal:** None.

Poster

PSTR136. Neuroinflammation: HIV, HSV, COVID19, and Other Infections I

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR136.04/I7

Topic: C.02. Alzheimer's Disease and Other Dementias

Title: Using Natural Language Processing to identify psychiatric symptoms related to Alzheimer's disease: a novel tool to enhance molecular brain research in Alzheimer's disease

Authors: *W. JIANG[1], J. VOGELGSANG[1], S. DAN[2], S. BERRETTA[3], T. KLENGEL[1]; [1]McLean Hosp., Belmont, MA; [2]McLean Hospital, HMS, Belmont, MA; [3]Translational Neurosci. Lab., Mrc3- McLean Hosp., Belmont, MA

Abstract: Neuropsychiatric symptoms (NPS), such as anxiety, depression, or agitation, are common across different stages of Alzheimer's disease (AD). However, their molecular correlates are poorly understood. While post-mortem brain research provides a unique source to study brain-specific molecular changes, clinical characterization of post-mortem brain donors is challenging and primarily subject to case-control designs. Here, we present a novel approach to post-mortem brain donor characterization using natural language processing (NLP).We analyzed electronic health records (EHR) of 92 brain donors (46 males, 46 females, mean age 81.9 years, Braak & Braak stages 0 - VI) using NLP-based NIMH Research Domain Criteria (RDoC) quantification. Individual RDoC scores were regressed to their neuropathological findings and bulk RNAseq data from two independent brain regions, the anterior cingulate gyrus and insula.Neuritic plaques (NP) in all four cortices show a high association with RDoC "cognition", controlled for age and sex (frontal: ß = 0.38, $p < 0.001$; parietal: ß = 0.35, $p < 0.001$; temporal: ß = 0.37, $p < 0.001$; occipital: ß = 0.37, $p < 0.001$). These findings align with prior clinicopathological correlation studies where cognitive examinations were performed within one year before death.At $p_{FDR}<0.05$ and $\log_2(\text{Fold Change})>0.58$, we identified 128 differentially expressed genes (DEGs) in the insula and 42 DEGs in the anterior cingulate gyrus between AD

and controls. We have further built a model regressing individual RDoC levels against their DEGs in the two brain regions. In each domain, we observed a cluster of genes that is up- or downregulated in relation to their RDoC score (higher/lower domain burden). In addition, we show that the majority of DEGs are RDoC-specific. 223 DEGs were uniquely related to one domain (170 DEGs in the insula and 53 DEGs in the anterior cingulate gyrus), while a total of 211 DEGs were associated with at least two domains (151 DEGs in the insula and 60 DEGs in anterior cingulate gyrus).NLP-based analysis of RDoC can be applied to post-mortem case collections to phenotype brain donors and show promising associations with neuropathology or RNAseq data that can be used to discover novel molecular pathways related to psychiatric symptoms.

Disclosures: W. Jiang: None. **J. Vogelgsang:** None. **S. Dan:** None. **S. Berretta:** None. **T. Klengel:** None.

Poster

PSTR136. Neuroinflammation: HIV, HSV, COVID19, and Other Infections I

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR136.05/I8

Topic: C.07. Neurotoxicity, Inflammation, and Neuroprotection

Support: NIH Grant K08NS119882

Title: Hexb modulates susceptibility to virally induced prenatal neuronal injury

Authors: *L. TRAN[1,2], L. HENDERSON[2], A. NATH[2], Y. KOUSA[1,2];
[1]Ctr. for Genet. Med. Res., Children's Natl. Med. Ctr., Washington, MD; [2]Natl. Inst. of Neurolog. Disorders and Stroke, NIH, Bethesda, MD

Abstract: The factors influencing diverse neurodevelopmental outcomes during prenatal viral infection remain poorly understood. Viral infections during this critical period can lead to severe neurodevelopmental disorders, though modifiers of brain injury are unknown. We hypothesize that differences in prenatal gene expression modulate susceptibility to virally induced brain injury.
Preliminary clinical data identified a candidate gene, *HEXB*, as a modifier for congenital Zika syndrome. *HEXB* encodes an enzyme responsible for degrading GM2 gangliosides, playing a vital role in lysosomal function and neuronal survival. *HEXB* is also implicated in autophagy—a host-defense catabolic process that traps and degrades viral cargo, like Zika, within lysosomes. Interestingly, closely-related viruses can exploit autophagic machinery to promote viral replication.
To characterize the relationship between Zika infection, *HEXB*, and neuronal injury, we used wildtype, heterozygous, and knockout *HEXB* induced pluripotent stem cells (iPSCs) to create an allelic series of neural stem cells (NSCs) and cerebral organoids. Monolayer neural stem cells were infected with Zika and collected at time points spanning early, mid, and late infection and

autophagy activation (12, 24, 36 hours). Autophagic flux and viral replication were quantified via RT-qPCR and Western Blotting. Four-week-old organoids were also infected to elucidate the impact of *HEXB* on cerebral development in a viral context. Furthermore, bulk RNA sequencing was performed to analyze transcriptional changes in the organoids 48 hours post-infection.
Our results reveal that *HEXB* is necessary for autophagy and acts as a viral restriction factor in neuronal development. Loss of *HEXB* leads to disrupted autophagy-related gene expression, including *p62, LC3B, WDR45,* and *NBR1*. Notably, both heterozygous and knockout *HEXB* mutants showed drastically increased levels of Zika viral replication compared to the wildtype counterpart. *HEXB* mutant NSCs also showed increased levels of cell death post-infection, suggesting that *HEXB* is required for neural stem cell survival. Infection of *HEXB*-mutated cerebral organoids results in significant size reduction relative to the wildtype. Bulk RNA-seq data show exacerbated disruption of autophagy-related genes.
Together, these findings suggest that loss-of-function mutations in *HEXB* can modulate susceptibility to prenatal virally induced neuronal injury. Understanding the molecular mechanisms involved in genetic susceptibility to viral infection paves the way for future targeted therapies and preventative strategies for these neurodevelopmental disorders.

Disclosures: **L. Tran:** None. **L. Henderson:** None. **A. Nath:** None. **Y. Kousa:** None.

Poster

PSTR136. Neuroinflammation: HIV, HSV, COVID19, and Other Infections I

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR136.06/J1

Topic: C.07. Neurotoxicity, Inflammation, and Neuroprotection

Support: NIH R01 DA052209
NIH R01 MH087332
NIH P50 DA026306 (P5)

Title: Methamphetamine exposure has long-lasting effects on expression of innate immune genes in the brain of a transgenic NeuroHIV model

Authors: *M. KAUL[1], R. SHAH[1], R. MAUNG[1], D. OJEDA-JUAREZ[1], I. HARAHAP-CARRILLO[1], A. J. ROBERTS[2], A. B. SANCHEZ[3], T. GROUP[4];
[1]SOM, Div. Biomed. Sci., Univ. of California Riverside, Riverside, CA; [2]Animal Models Core, The Scripps Res. Inst., La Jolla, CA; [3]Sanford Burnham Prebys Med. Discovery Inst., La Jolla, CA; [4]Dept. of Psychiatry, Univ. of California San Diego, La Jolla, CA

Abstract: Use of methamphetamine (METH) by people living with HIV-1 appears to increase the risk of developing HIV-associated neurocognitive disorders (HAND). Yet, the underlying pathological mechanisms are incompletely understood. Key neuropathological features of NeuroHIV/AIDS patients are recapitulated in transgenic mice expressing soluble viral envelope protein gp120 of HIV-1 in the brain (gp120tg). Previously, we exposed 4 months-old gp120tg

mice to an escalating METH binge regimen for 25 days and analyzed the animals 7 months afterwards. Both gp120 expression and METH exposure diminished hippocampal long-term potentiation but METH-exposed gp120tg animals also showed reduced post-tetanic potentiation. METH and gp120 exposure also caused lasting differential expression of neurotransmission-related genes, neuronal injury and behavioral impairment. In this study, we analyzed expression of genes linked to innate immunity, neuroinflammation and degeneration in cerebral cortex that have been implicated in the pathological mechanisms of NeuroHIV. The presence of viral gp120 caused increased expression of innate immune genes, such as IFNa, IFNb, IFNAR1, CCR5, LCN2, LTC4S, CysLTR1, and BACE. However, previous METH exposure resulted in a diminished induction or reduction for CCR5, LCN2 and BACE, and in a sex-dependent fashion for IFNAR1 and IFNb in females and CysLTR1 in males. On the other hand, METH triggered in males an increase of IFNa in the presence and absence of gp120, and of IFNAR1 in the presence of gp120. The expression of several of the innate immune and proinflammatory genes correlated significantly with that of astrocytic and microglial markers, but IFNAR1 also with neuronal Calb1 and Pvalb in males. An increase of IFNa in cerebral cortex and correlation of IFNAR1 with markers of inhibitory neurons is in line with a relevant effect of the cytokine on neurocognitive function. Altogether, METH and HIV affect innate immune and inflammatory factors in a lasting and partly sex-dependent fashion, and our study provides a framework for the identification of gene networks that play causal roles in neurocognitive impairment.

Disclosures: M. Kaul: None. **R. Shah:** None. **R. Maung:** None. **D. Ojeda-Juarez:** None. **I. Harahap-Carrillo:** None. **A.J. Roberts:** None. **A.B. Sanchez:** None. **T. Group:** None.

Poster

PSTR136. Neuroinflammation: HIV, HSV, COVID19, and Other Infections I

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR136.07/J2

Topic: C.07. Neurotoxicity, Inflammation, and Neuroprotection

Support: NIH
The University of Texas Rio Grande Valley
RGV-SFN-Chapter

Title: Utilizing Fluorescent Nanodiamonds for Targeted Drug Delivery TowardMicroglial Cells.

Authors: *E. OWUSU[1], D. ROY[1], S. MITO[2], M. ZAREI[1], E. A. ROZHKOVA[3], U. ROY[1]; [2]Dept. of Med. Educ., [1]The Univ. of Texas Rio Grande Valley, Brownsville, TX; [3]Ctr. for Nanoscale Materials, Argonne, IL

Abstract: Combination antiretroviral therapy (cART) has been accepted for the treatment of HIV infection across the globe. However, there is a limitation to this treatment as it cannot successfully reach viral reservoir cells such as microglial in the brain. Therefore, a significant number of HIV infected patients develop HIV-associated neurological disorder (HAND). Recent

advancement in nanomedicine-based targeted drug delivery has provided the opportunity to deliver drug that target microglial cells. To address this issue, researchers have been exploring nanodiamond (ND)-based drug delivery systems that target microglia. NDs have gained attention due to their natural biocompatibility, non-toxicity, and efficiency as nano-carriers, making them a promising option over the other carbon-based nanomaterials. Building on our previous studies that demonstrated the efficacy of NDs crossing the blood brain barrier (BBB), our current study aims to enhance this approach using fluorescent nanodiamonds (FNDs). Although FNDs are like NDs in their nano size, large surface area available for attachment of biological molecules, and high colloidal stability, these nanostructures have an important additional property of traceability. In our study, we have developed a multifunctional, traceable nanodrug platform specifically designed to transmigrate across BBB tight junctions and target microglial viral reservoirs in the brain. Our various *in vitro* experiments such as cytotoxicity assay, detection of reactive oxygen species (ROS), immunocytochemical assays using human microglial cell line (HMC-3) have proven that FNDs are competent and biocompatible to deliver drugs to the brain. Our *in vitro* cytotoxicity data indicated that FNDs were non-toxic to HMC-3. The *in vitro* immunocytochemistry data also showed the FNDs were taken up by the microglia without any cytotoxic effect. We have further hypothesized that the drug payload will be released and subsequently taken up by resident microglia once they enter the brain. This is expected to lead to a reduction in viral load in the brain, which will significantly improve neurological pathology and therefore improve the lives of people with HIV (PWH).

Disclosures: **E. Owusu:** None. **D. Roy:** None. **S. Mito:** None. **M. Zarei:** None. **E.A. Rozhkova:** None. **U. Roy:** None.

Poster

PSTR136. Neuroinflammation: HIV, HSV, COVID19, and Other Infections I

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR136.08/J3

Topic: C.07. Neurotoxicity, Inflammation, and Neuroprotection

Support: NIH NIDA Grant DA054992
NIH NIDA Grant DA058586

Title: Suppression of HIV and cocaine-induced neurotoxicity and inflammation by cell penetrable esters of itaconate

Authors: M. AKSENOVA[1], B. CUI[1], L. KORUNOVA[1], A. SIKIRZHYTSKAYA[1], V. SIKIRZHYTSKI[1], H. JI[1], E. BROUDE[1], M. D. WYATT[1], N. FRIZZELL[2], R. BOOZE[3], ***M. SHTUTMAN**[1];
[1]Drug Discovery and Biomed. Sci., Univ. of South Carolina, Columbia, SC; [2]Pharmacology, Physiology, and Neurosci., Univ. of South Carolina Sch. of Med., Columbia, SC; [3]Psychology, Univ. South Carolina, Columbia, SC

Abstract: Suppression of HIV and cocaine–induced neurotoxicity and inflammation by cell penetrable estersof itaconate

HIV Associated Neurocognitive Disorder (HAND) is one of the most common complications of HIV infection. There is an urgent need for effective therapeutic strategies for HAND exacerbated by Cocaine Use Disorder (CUD). Through an extensive analysis of the HIV-induced transcriptome, we determined that HIV and cocaine profoundly induce overexpression of the microglia-specific gene aconitate decarboxylase 1 (acod1). Acod1 converts the TCA intermediate cis-aconitate to itaconate during macrophage inflammation. Although the attenuation of inflammation by itaconate has been characterized in peripheral macrophages, the role of immunometabolism, including itaconate production, has not been studied in HIV and cocaine-exposed microglia. BBB penetrable ester modifications of itaconate, 4-octyl-itaconate (4-OI), have a potent anti-inflammatory activity and have been proposed as promising medicines for a range of diseases. We tested the activity of the 4-OI in HIV-Tat and cocaine-treated rat primary neuronal cultures and found a protective effect on viability of 4-OI against the combined toxicity of HIV-Tat and cocaine. Since Acod1 is microglia-specific, we then determined the effects of 4-OI on microglia. Tat and cocaine treatment profoundly increased the body size of microglia and decreased the length of their processes, the "classical" phenotype for pathological activation. 4-OI increased the number of microglial cells in both control and HIV-Tat/cocaine-treated primary cortical cultures, and reversed the HIV-Tat and cocaine-induced morphological changes. In the presence of 4-OI, microglial cells appeared more ramified, like untreated microglia. Corroborating these results, 4-OI treatment also inhibited the secretion of the proinflammatory cytokines IL1α, IL-1β, IL6, MIP1-α induced by HIV-Tat and cocaine. Transcriptome profiling further determined that the Nrf2 target genes NQO1, Gstp1, and GCLc are most activated in 4-OI treated primary cortical cultures after Tat insult, relative to Tat alone. These genes are well established regulators of anti-inflammatory response that inhibit ROS production, reduce the secretion of pro-inflammatory cytokines, and suppress NF-kB signaling. The expression of genes regulating cytoskeletal dynamics in inflammatory microglia are downregulated by 4-OI treatment. The results suggested 4-OI as a promising candidate for development of therapeutics for the treatment of HAND with CUD comorbidities.

Disclosures: M. Aksenova: None. **B. Cui:** None. **L. Korunova:** None. **A. Sikirzhytskaya:** None. **V. Sikirzhytski:** None. **H. Ji:** None. **E. Broude:** None. **M.D. Wyatt:** None. **N. Frizzell:** None. **R. Booze:** None. **M. Shtutman:** None.

Poster

PSTR136. Neuroinflammation: HIV, HSV, COVID19, and Other Infections I

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR136.09/J4

Topic: C.07. Neurotoxicity, Inflammation, and Neuroprotection

Support: NIH Grant Award DA046258

Title: Network meta-analysis of the molecular mechanisms and signal pathways underlying augmentation of depression in the course of COVID-19

Authors: R. PATEL[1,2], J. ZHANG[1,2], M. BISHIR[1,2], H.-H. CHEN[3], S. CHANG[1,2];
[1]Inst. of NeuroImmune Pharmacol., South Orange, NJ; [2]Dept. of Biol. Sci., Seton Hall Univ., South Orange, NJ; [3]Natl. Hlth. Res. Inst., Zhunan,, Taiwan

Abstract: SARS-Cov-2 infection causes COVID-19 that has affected over 750 million people and is responsible for almost 7 million deaths according to the World Health Organization. COVID-19 is marked by a cytokine storm characterized by an influx of inflammatory molecules. The cytokine storm has been found to facilitate the development of neuroinflammation, however the mechanisms behind the facilitation have not been studied in depth. Furthermore, there is an increased prevalence of psychological disorders, especially depression, during and after COVID-19 infection. Many environmental factors that influence the development of depression have been suggested, but biological factors are not studied comprehensively. Elevated systemic inflammation leading to neuroinflammation has been reported prior to the development of neuronal abnormality possibly including depression. With these premises, we have hypothesized that neuroinflammation induced by COVID-19 cytokine storm can lead to augmentation of depression. QIAGEN Ingenuity Pathway Analysis (IPA) tools were used to investigate the molecular mechanisms and signaling pathways underlying how cytokine storm induced neuroinflammation modulates the development of depression in COVID-19. In parallel, two GSE datasets collected from GEO, GSE157103 and GSE18884, were analyzed using GEO2R to identify differentially expressed genes (DEGs) with COVID-19. GSE157103, a study on "Large-Scale Multi-omic Analysis of COVID-19 Severity" was used to examine COVID-19's effects on plasma and leukocytes to reveal molecular mechanisms underlying the cytokine storm and neuroinflammation. GSE188847, a study on "Severe COVID-19 is associated with molecular signatures of aging in the human brain," was used to examine COVID-19's effect on the frontal cortex (FC) to reveal signaling pathways underlying depression. IPA's Core Analysis of the COVID-19-induced DEGs in leukocytes revealed 1) the inhibition of EIF2 and IL-15 Signaling pathways and 2) the activation of p70s6k and B Cell Development Pathways suggesting the development of inflammation. The parallel analysis of COVID-19-induced DEGs in the FC reveals the inhibition of 1) Synaptogenesis, 2) Serotonin Receptor, and 3) α-Adrenergic Signaling Pathways suggesting augmentation of depression. Finally, the COVID-19-induced DEGs were overlaid onto the network pathways established using QKB to demonstrate the overall augmentation of depression. Taken together, our network meta-analyses provided empirical evidence for the augmentation of depression during COVID-19 and possible post-COVID-19 pandemic.

Disclosures: R. Patel: None. **J. Zhang:** None. **M. Bishir:** None. **H. Chen:** None. **S. Chang:** None.

Poster

PSTR136. Neuroinflammation: HIV, HSV, COVID19, and Other Infections I

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR136.10/J5

Topic: C.07. Neurotoxicity, Inflammation, and Neuroprotection

Title: Understanding HSV-1 host miRNA-mediated mechanisms of post-transcriptional gene regulation in the CNS

Authors: *B. H. POWELL[1], Z. LIAO[2], C. MONTEIRO ABREU[2], W. T. MILLS, IV[3], P. DESAI[4], M. K. MEFFERT[3], K. W. WITWER[5];
[1]Biol. Chem. and Mol. & Comparative Pathobiology, [2]Mol. & Comparative Pathobiology, [3]Biol. Chem., [4]Oncology, Viral Oncology, [5]Mol. & Comparative Pathobiology and Neurol., Johns Hopkins Univ., Baltimore, MD

Abstract: Herpes Simplex Virus 1 (HSV-1) infects 50-80% of the United States population. HSV-1 establishes latency in sensory neurons for the host's lifetime but can sporadically reactivate. In some instances, reactivated HSV-1 can enter the central nervous system and cause lethal disorders. Latency and reactivation are controlled by pronounced changes in expression of the viral genome, with latency characterized by lifelong retention of the HSV-1 genome in a predominantly 'silent' state with the exception of one highly expressed region encoding the Latency Associated Transcript (LAT). The HSV-1 genome encodes 18 precursor microRNAs (miRNAs). Both host and HSV-1 encoded miRNAs are implicated in post-transcriptional regulatory roles governing state transitions between HSV-1 latency and reactivation. However, the gene regulatory interactions and molecular mechanisms remain incompletely characterized. This study evaluates regulatory mechanisms of host and HSV-1 encoded miRNA interactions with target RNAs during latency in infected human neurons. The functional roles of miRNA:target RNA interactions are investigated, including through testing the effects of global inhibition of miRNA-mediated repression in HSV-1 human excitatory neurons derived from induced pluripotent stem cells (iPSCs). The effects of disrupting implicated miRNA regulatory nodes are analyzed to test the functional importance of specific interactions in governing HSV-1 latency or reactivation. Insight into miRNA-mediated regulation of gene expression and state transitions in HSV-1 infected cells has the potential to aid in developing novel approaches to treat and prevent HSV-1 reactivation.

Disclosures: B.H. Powell: None. **Z. Liao:** None. **C. Monteiro Abreu:** None. **W.T. Mills:** None. **P. Desai:** None. **M.K. Meffert:** None. **K.W. Witwer:** None.

Poster

PSTR136. Neuroinflammation: HIV, HSV, COVID19, and Other Infections I

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR136.11/J7

Topic: C.07. Neurotoxicity, Inflammation, and Neuroprotection

Support: R01-NS104016
R21-MH125716

P51-OD11104
U42-OD024282
U42-OD010568

Title: Astrocyte expression of cellular senescence marker $p16^{INK4a}$is significantly increased in the frontal lobe, hippocampus, thalamus, caudate, and cerebellum following SIV infection: Implications for HIV-associated neurocognitive disorders in cure research

Authors: *M. HORN[1,2], A. G. MACLEAN[2,3,4,5];
[2]Brain Inst., [1]Tulane Univ., New Orleans, LA; [3]Div. of Comparative Pathology, Tulane Natl. Primate Res. Ctr., Tulane Univ., Covington, LA; [4]Dept. of Microbiology & Immunology, Sch. of Med., [5]Ctr. for Aging, Tulane Univ., New Orleans, LA

Abstract: Prior to the use of combination antiretroviral therapies (cART) in the treatment of HIV, the lifespan of people living with HIV (PLWH) was over a decade shorter than HIV-negative individuals. While cART largely closed this gap, it has had seemingly no effect on the gap in healthspan, or time without comorbidity, which remains around a decade shorter for PLWH. As many of these comorbidities are age-related - including heart disease, diabetes, and neurocognitive impairments - it is hypothesized that PLWH experience premature or accelerated aging. This research aims to determine the extent of premature aging in the CNS of Simian immunodeficiency virus (SIV) infected rhesus macaques and assess the efficacy of several therapeutic strategies in limiting or reversing these effects. Based on the prevalence of HIV-associated neurocognitive disorders (HAND) remaining around 50% despite the advent of cART, we hypothesized that exposure to HIV/SIV increases the rate of biological aging, and that cART has little to no effect on the extent of premature or accelerated aging in the CNS. To test this hypothesis, we have applied an immunofluorescence assay, whole slide imaging, and HALO® image analysis to archival FFPE tissues from naïve (n = 3), SIV-infected (SIV, n = 5), and SIV-infected, cART-treated (SIV-cART, n = 7) rhesus macaques of both sexes, ranging from four to fifteen years of age. We analyzed the percentage of cells expressing the cellular senescence marker $p16^{INK4a}$/CDKN2A (p16) in multiple brain regions implicated in HAND, including the frontal lobe, caudate, putamen, hippocampus, thalamus, and cerebellum. Compared to naïve animals, SIV and SIV-cART animals had significantly higher percentages of total p16+ cells in the frontal lobe (SIV $p < 0.001$, SIV-cART $p < 0.001$), hippocampus (SIV $p < 0.001$, SIV-cART $p = 0.0048$), thalamus (SIV $p = 0.0040$, SIV-cART $p < 0.0001$), and cerebellum (SIV $p = 0.0032$, SIV-cART $p = 0.0033$), but no difference in either the caudate or putamen. When we assessed colocalization of the astrocytic marker GFAP with p16, in comparison to naïve animals, we found a significant increase in the percentage of double-labeled cells in the frontal lobe (SIV $p < 0.001$, SIV-cART $p < 0.001$), hippocampus (SIV $p < 0.001$, SIV-cART $p = 0.0019$), thalamus (SIV $p = 0.0011$, SIV-cART $p < 0.001$), cerebellum (SIV $p < 0.001$, SIV-cART $p < 0.001$) and caudate (SIV-cART $p = 0.0392$). While p16 expression alone is insufficient to indicate premature or accelerated aging, this data suggests exposure to HIV/SIV may induce cellular senescence in astrocytes, contributing to the continued prevalence of HAND in the post-cART era and warrants further investigation.

Disclosures: M. Horn: None. **A.G. MacLean:** None.

Poster

PSTR136. Neuroinflammation: HIV, HSV, COVID19, and Other Infections I

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

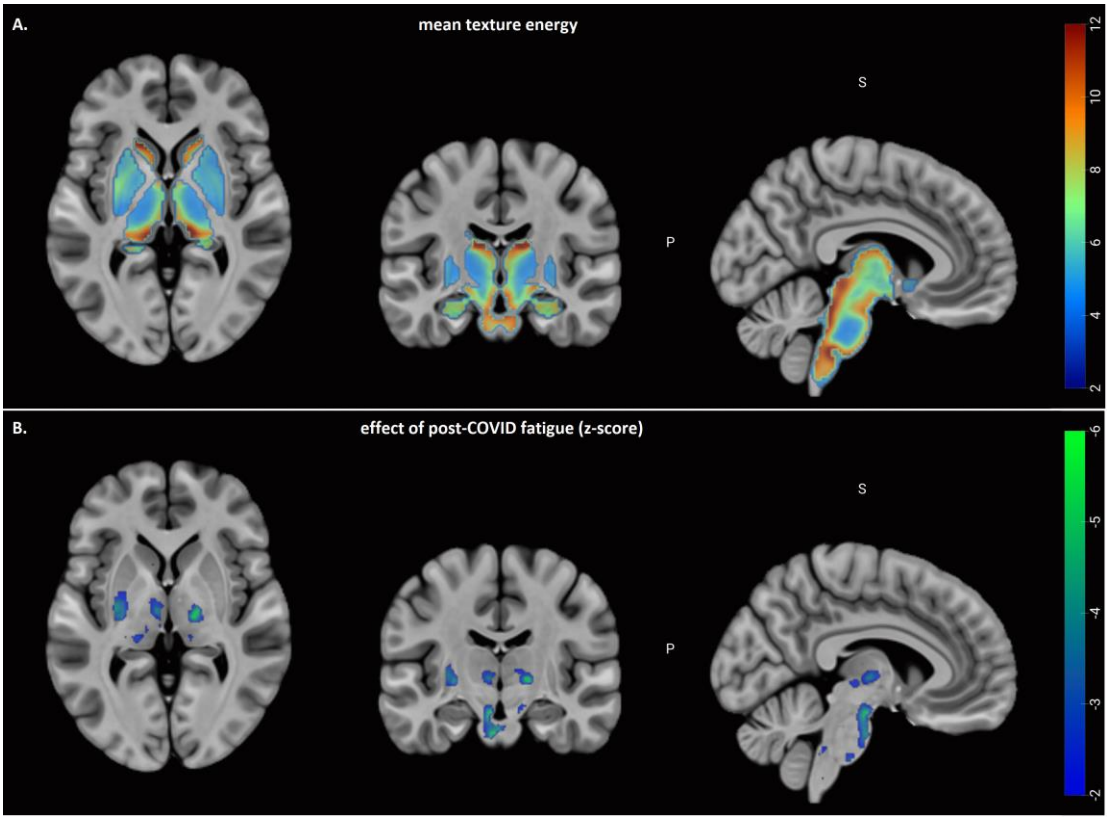
Program #/Poster #: PSTR136.12/J8

Topic: C.07. Neurotoxicity, Inflammation, and Neuroprotection

Title: Post-covid fatigue is associated with abnormal subcortical texture in T1-weighted MRI

Authors: *N. CHURCHILL[1], E. ROUDAIA[2], J. J. CHEN[3], A. B. SEKULER[3], F. GAO[4], M. MASELLIS[5], B. LAM[4], I. CHENG[4], C. HEYN[4], S. E. BLACK[6], B. J. MACINTOSH[4], S. J. GRAHAM[7], T. A. SCHWEIZER[8];
[1]St. Michael's Hosp., Toronto, ON, Canada; [2]Rotman Res. Inst., Montreal, QC, Canada; [3]Baycrest Hosp., Toronto, ON, Canada; [4]Sunnybrook Hosp., Toronto, ON, Canada; [5]Sunnybrook Res. Inst., Toronto, ON, Canada; [6]Dept Med. (Neurol), Sunnybrook Hlth. Sci. Cntr, Toronto, ON, Canada; [7]Research, Physical Sci., Sunnybrook Hlth. Sci. Ctr., Toronto, ON, Canada; [8]Univ. of Toronto, Toronto, ON, Canada

Abstract: The coronavirus disease 2019 (COVID-19) represents an unprecedented public health crisis. These is also growing evidence that the disease affects the central nervous system, via both direct and indirect pathways. These effects may be long-lasting, with growing case numbers of post-acute COVID-19 syndrome (PACS), in which symptoms and neurological issues persist more than 12 weeks post-infection. Among the symptoms associated with PACS, fatigue is a major concern, given its high prevalence and impact on daily functioning. In other disorders with high rates of fatigue, subcortical structures are frequently implicated, such as the brainstem, basal ganglia and thalamus. These areas are also vulnerable to COVID-related injury, identified via clinical imaging and neuropathology. Hence, imaging biomarkers of microstructural injury that correlate with post-COVID fatigue are of significant interest. A promising approach to this challenge is texture-based analysis (TBA) of structural brain scans. This approach can provide information about subtle changes in tissue microstructure that cannot be discerned by eye. In the present study, we apply TBA to subcortical structures of T1-weighted anatomical scans collected as part of the Toronto-based NeuroCOVID-19 study. We compared Haralick texture features for self-isolating individuals who tested positive for SARS-CoV-2 and had persistent symptoms, along with controls who had cold or flulike symptoms but tested negative for SARS-CoV-2, with both groups imaged an average of 4-5 months after COVID testing. Significant differences were seen in grey matter texture for COVID-19 patients with persistent fatigue, relative to both COVID-19 patients without fatigue and controls. This included decreased Energy (-15.0% ± 2.1%) and increased Entropy (7.9% ± 1.0%) in the brain stem and thalamus, along with decreased Correlation (-25.3% ± 4.1%) in the putamen, all at $p<0.001$. These findings provide encouraging evidence for abnormal tissue texture as a biomarker of post-COVID fatigue, providing new insights into this highly prevalent disorder.



Disclosures: **N. Churchill:** None. **E. Roudaia:** None. **J.J. Chen:** None. **A.B. Sekuler:** None. **F. Gao:** None. **M. Masellis:** None. **B. Lam:** None. **I. Cheng:** None. **C. Heyn:** None. **S.E. Black:** None. **B.J. Macintosh:** None. **S.J. Graham:** None. **T.A. Schweizer:** None.

Poster

PSTR136. Neuroinflammation: HIV, HSV, COVID19, and Other Infections I

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR136.13/J9

Topic: C.07. Neurotoxicity, Inflammation, and Neuroprotection

Support: CIHR Grant GA4-177756
Sunnybrook Foundation
Mitacs
Siemens Healthineers

Title: Brain Effects of Post-COVID-19 Condition Observed by Dynamic Susceptibility Contrast Magnetic Resonance Imaging

Authors: ***S. J. GRAHAM**[1], A. PAVEL[1], F. O'HARA[1], N. W. CHURCHILL[3], F. TAM[1], F. GAO[2], M. MASELLIS[2], B. LAM[2], I. CHENG[2], C. HEYN[2], E. ROUDAIA[4], J. CHEN[4], S. E.

BLACK[2], A. B. SEKULER[4], T. A. SCHWEIZER[3], B. MACINTOSH[2];
[1]Physical Sciences, Sunnybrook Res. Inst., [2]Hurvitz Brain Sci. Program, Sunnybrook Res. Inst., Sunnybrook Hlth. Sci. Ctr., Toronto, ON, Canada; [3]St. Michael's Hospital, Unity Hlth. Syst., Toronto, ON, Canada; [4]Rotman Res. Inst., Baycrest, Toronto, ON, Canada

Abstract: **Research Objective** Post-coronavirus disease 2019 condition (PCC) is prevalent, with high socioeconomic and healthcare burden internationally. Many common PCC symptoms suggest brain injury, but the underlying biological mechanisms remain poorly understood. Towards filling this knowledge gap and developing targeted brain treatments, the NeuroCOVID19 study commenced in spring 2020 involving detailed magnetic resonance imaging (MRI) of the brain; electroencephalography; and assessment of symptoms and behaviors[1]. Notably, dynamic susceptibility contrast (DSC) MRI is included to assess cerebral microvessel pathophysiology [2], and test whether DSC cerebral blood flow (CBF) and vascular leakage parameter K2 are altered in PCC individuals compared to controls. **Methods** To date, DSC data have been collected for 14 healthy controls (9 female (F), mean (standard deviation) age = 44(13)), and 63 PCC participants (55 self-isolated while infected, 37 F, age = 42(12); 8 hospitalized while infected, 5 F, age=54(11)). DSC MRI was performed at 3 T (1.74 mm in-plane resolution, 8 mm slices, 140 time points, 1.25 s temporal resolution) using Gadovist contrast agent. Data were analyzed with a custom pipeline to generate CBF and K2 maps (see representative healthy control data, Fig. 1) followed by voxel-wise two-tailed t-tests with correction for multiple comparisons.**Results/Conclusions** No statistically significant group differences were observed, but there were notable trends between self-isolated PCC individuals and controls for CBF and K2 when a region of interest analysis was conducted for the thalamus - consistent with previous NeuroCOVID19 CBF results obtained using arterial spin labeling[3], and with reports that leaky brain vasculature elevates risk of acute COVID19[1]. These results help confirm that there are observable changes in cerebrovascular physiological parameters due to PCC, either as a direct or indirect consequence of SARS-CoV2 infection.1. Macintosh BJ et al., CMAJ Open 2021.2. Boxerman JL et al., AJNR 2006.3. Kim WSH et al., JMRI 2022.

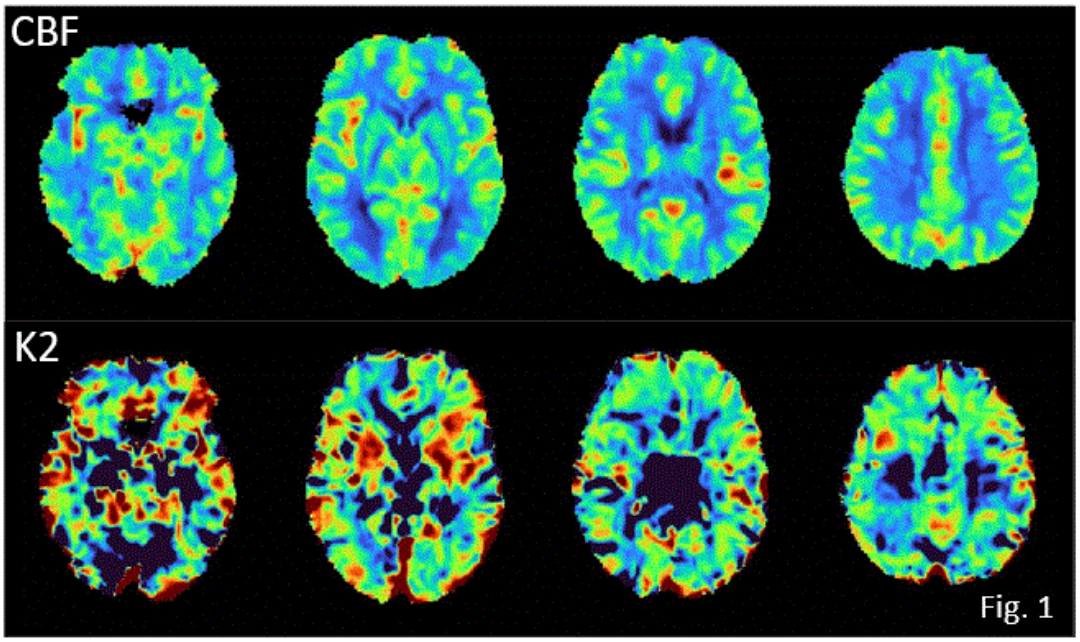


Fig. 1

Disclosures: **S.J. Graham:** None. **A. Pavel:** None. **F. O'Hara:** None. **N.W. Churchill:** None. **F. Tam:** None. **F. Gao:** None. **M. Masellis:** None. **B. Lam:** None. **I. Cheng:** None. **C. Heyn:** None. **E. Roudaia:** None. **J. Chen:** None. **S.E. Black:** None. **A.B. Sekuler:** None. **T.A. Schweizer:** None. **B. Macintosh:** None.

Poster

PSTR136. Neuroinflammation: HIV, HSV, COVID19, and Other Infections I

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR136.14/J10

Topic: C.07. Neurotoxicity, Inflammation, and Neuroprotection

Support: R01 DA045596
R21 DA057871

Title: Memory Assessment at Single Cell Resolution Using In vivo Calcium Imaging in HIV-1 Tat Transgenic Mice Exposed to Δ^9-Tetrahydrocannabinol

Authors: *B. J. YADAV-SAMUDRALA[1], R. PATEL[2], S. FITTING[3];
[1]Psychology and Neurosci., Univ. of North Carolina Chapel Hill, Chapel Hill, NC; [2]Neurosci. Ctr., Univ. of North Carolina at Chapel Hill, Chapel Hill, NC; [3]Psychology & Neurosci., Univ. of North Carolina, Chapel Hill, NC

Abstract: **Background:** Despite the advancement of combined anti-retroviral therapies (cART) the prevalence of HIV-1-associated neurocognitive disorders (HAND) remains high. Cannabis use is known to alleviate common symptoms and complications in people living with HIV (PLWH) on cART but its effect on neurocognition is not clear. With the use of *in vivo* calcium imaging this study aims to determine cannabis-induced changes on neuronal activity in HIV-1 Tat transgenic (tg) mice during performance of a memory-related novel object recognition (NOR) task. **Methods:** HIV-1 IIIB Tat_{1-86} tg male mice (~2-3 months old, $N = 15$) were microinjected with GCaMP6f in the prefrontal cortex (PFC) (coordinates: +1.87 A/P, -0.5 M/L, -2.4 D/V). A ProView Integrated lens (Inscopix) was implanted over the injection site & secured in place with metabond. Control Tat(-) mice ($n = 6$) and Tat(+) mice ($n = 7$) were assessed in the NOR task at 8 weeks post-surgery in the presence of vehicle and Δ^9-tetrahydrocannabinol (THC, 10 mg/kg). Data was collected using the nVista system (Inscopix), and behavioral recordings were synchronized on the e3vision camera (White Matter) with the 20 Hz recordings of the miniscope (triggered with a 5V TTL pulse). Calcium transient time series were analyzed through the pipelines created by Inscopix. **Result:** We successfully established the *in vivo* calcium imaging technique in our HIV-1 Tat tg mouse model with imaging >1000 individual neurons for each genotype. Whereas no genotype differences were noted for locomotor activity, acute 10 mg/kg THC injections downregulated overall object exploration behavior in all mice. *In vivo* calcium imaging of PFC cellular activity during the NOR task revealed a correlation between neuronal activity and behavior for control Tat(-) mice. Specifically, when treated with vehicle, neuronal activity was highly associated with exploration of the familiar object, whereas 10

mg/kg THC reversed neuronal activity to be highly correlated with exploration of the novel object. No association between neuronal activity and behavior was noted for Tat(+) mice. Additional data analyses for event detection at a single cell resolution is currently underway. **Conclusion:** Our data suggests that PFC network activity in HIV Tat tg mice is altered by genotype and THC exposure during performance of a memory-related behavioral task.

Disclosures: B.J. Yadav-Samudrala: None. **R. Patel:** None. **S. Fitting:** None.

Poster

PSTR137. Spinal Cord Injury—Therapeutic Strategies In Vivo: Pharmacological

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR137.01/K1

Topic: C.11. Spinal Cord Injury and Plasticity

Support: Supported by Merit Review Award # B3986-R/1 I01 RX003986-01A1, from the United States Department of Veterans Affairs Rehabilitation Research and Development Service (RR&D)
Spinal Cord Injury Research Program (SCIRP) Investigator-Initiated Research Award # SC210266 from the United States Department of Defense (DoD)

Title: Iron chelator therapy improved hallmark spinal cord injury (SCI) disabilities by reducing hemorrhagic iron toxicity, inflammation, and pyroptosis in a rodent model of cervical SCI

Authors: *P. BOSE[1,2,3], J. HOU[1,2], S. TSUDA[1,2], D. PLANT[1], G. M. DOOLEY[2,1], K. S. KLIPPEL[2,1], G. HWANG[2,1], G. A. VARGAS[2], H. SADEESHKUMAR[1], R. J. BERGERON, Jr[4], F. J. THOMPSON[1,5];
[1]North Florida/South Georgia Veterans Hlth. Syst., Gainesville, FL; [2]Anesthesiol., [3]Neurol., [4]Medicinal Chem., [5]Neurosci., Univ. of Florida, Gainesville, FL

Abstract: Cervical spinal cord injury (C-SCI) is a common and frequently devastating injury that can result in a broad range of life-long locomotor and spasticity disabilities. With advances in early evacuation and aggressive medical therapy, there are still no effective therapeutics that salvage spinal cord (SC) neurons and or reduce progressive secondary damage. Acceleration/deceleration and contusion SCI cause micro-vessel shear injury, blood spinal cord barrier (BSCB) dysfunction, and hemorrhage. Iron deposited by diffuse micro-hemorrhage fuels oxidative stress and inflammation through reactive oxygen species (ROS), which may further induce progressive disabilities. There is an urgent need to address both specific disabilities and risk factors for long-term progressive disabilities and to develop effective therapies that have excellent potential for translation. This study was aimed at testing the preclinical evaluation of the safety and efficacy of a new iron chelator, SP420, in a rodent model of contusion C-SCI. SP420 was administered SQ (66 mg/kg; represents the human phase II dose) in both acute and chronic time points and tested against saline placebo controls. Treatment was initiated 30

minutes after SCI and post-injury week-4 for 2 weeks, each using a separate cohort of animals. Quantitative physiological measures of spasticity, gait, and the integrity of axonal conduction of descending locomotor pathways functions were the primary outcomes, along with clinically relevant T1/T2W, SWI/QSM, and DTI MRIs. A comprehensive list of safety outcomes was applied during the treatment. A cause-effect relationship between iron deposition, tissue damage, and treatment effects of iron chelator was studied using a combination of histological and immunohistochemical assays to evaluate bleed iron, oxidative stress, inflammation, markers for BSCB integrity, and neural and vascular protective factors. Our data to date indicate that free bleed iron fuels oxidative stress and neuroinflammation through ROS, which, in part, drives the progression of neurological damage and motor disabilities. Our preliminary data also indicate that the SP-420 therapy reverses the iron-mediated neurological damage and delayed neurological sequelae in spasticity and gait disabilities. To amplify the robustness necessary to significantly improve function in a chronic setting of SCI, future studies will apply the combination of locomotor exercise and SP420 as complementary therapies. Achievement of these goals will provide innovative, non-invasive, and patient-centered technologies and treatments that will greatly facilitate the treatment of patients with SCI.

Disclosures: **P. Bose:** None. **J. Hou:** None. **S. Tsuda:** None. **D. Plant:** None. **G.M. Dooley:** None. **K.S. Klippel:** None. **G. Hwang:** None. **G.A. Vargas:** None. **H. Sadeeshkumar:** None. **R.J. Bergeron:** None. **F.J. Thompson:** None.

Poster

PSTR137. Spinal Cord Injury—Therapeutic Strategies In Vivo: Pharmacological

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR137.02/K2

Topic: C.11. Spinal Cord Injury and Plasticity

Support: 1R01EY033652

Title: Developing highly effective peptide drugs for promoting axon regeneration after CNS injury in adult rodents

Authors: *S. WANG[1,2,3], S. LI[1,2,3];
[1]Temple Univ., Philadelphia, PA; [2]Shriners Hosp. Pediatric Res. Ctr., Philadelphia, PA; [3]Dept. of Neural Sci., Philadelphia, PA

Abstract: Currently, there is no cure for patients with CNS axon injury and it is essential to develop highly effective therapeutic strategies for regenerating lesioned CNS axons. Targeting certain genes, including PTEN suppression, could stimulate strong regrowth of injured CNS neurons, but none of these gene targets have been translated to clinical trials for human treatments. It thus is vital to develop deliverable therapeutic strategies for promoting robust axon regeneration following CNS injuries. Previously, we designed five PTEN antagonist peptides (called PAPs 1-5) and reported some of them promoted significant regrowth of injured spinal

cord axons in adult mice [Biomaterials. 2014 May;35(16):4610-26]. By collaborating with Dr. Angelo Lepore's group at Jefferson University, we also verified the efficacy of PAP4 for treating acute and chronic cervical spinal cord injury in adult rats. However, the overall efficiency of our PAPs1-4 for promoting axon regrowth appears lower than that of transgenic PTEN deletion. Recently, we designed six more PAPs (called PAPs 6-9 and PAPLs1-2) by targeting the additional critical activity domains of PTEN protein. We evaluated and compared the efficacies of the 11 PAPs for promoting the regeneration of injured CNS axons using an optic nerve crush model in adult mice. Post-injury intravitreal treatments with each of these PAPs stimulated various degrees of optic nerve regeneration, but several of them, especially PAPL2, PAP3, and PAP9, promoted more robust regrowth and principally mimicked the effects of transgenic PTEN deletion. Similar to PTEN knockout, PAP treatments also significantly increased the survival of retinal ganglion cells in adult mice with optic nerve crushes. Our findings suggest the great potential of our small PTEN-blocking peptide drugs for treating neurological disorders characterized by CNS axonal injuries.

Disclosures: S. Wang: None. **S. Li:** None.

Poster

PSTR137. Spinal Cord Injury—Therapeutic Strategies In Vivo: Pharmacological

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR137.03/K3

Topic: C.11. Spinal Cord Injury and Plasticity

Support: VA-ORD 1I01BX005287

Title: Chemogenetic neuromodulation of cervical spinal interneurons: a novel preclinical translation strategy to restore breathing after chronic cervical spinal cord injury

Authors: *K. KONKEL, A. BREZINSKI, S. N. KURPAD, K. SATKUNENDRARAJAH; Med. Col. of Wisconsin, Milwaukee, WI

Abstract: Chemogenetic neuromodulation of cervical spinal interneurons: a novel preclinical translation strategy to restore breathing after chronic cervical spinal cord injury

Katherine Konkel[2,3], Allison Brezenski[1,2], Shekar Kurpad[2,3]1,2, Kajana Satkunendrarajah[2,3]

Cervical spinal cord injury (cSCI) impairs the descending drive to the spinal respiratory circuitry, leading to significant respiratory compromise. Despite mechanical ventilation, dysfunctional breathing remains a major cause of morbidity and mortality after cSCI. Restoring breathing in the chronic phase of cSCI is a challenging goal. Previous research has shown that stimulating a small population of cervical spinal excitatory interneurons (eINs) can acutely increase respiratory motor output. However, whether this strategy can be translated into a non-transgenic preclinical model to improve breathing in the late chronic phase of cSCI is unclear. To address this, we employed adeno-associated virus (AAV) technology to selectively express designer

receptors exclusively activated by designer drug (DREADD) in cervical eINs four weeks after C2 level spinal cord hemisection (C2Hx). Two weeks after viral injections, cervical eINs were stimulated by subcutaneously administering CNO, the DREADD ligand. Using electromyography, we evaluated the effect of CNO-mediated activation of cervical eINs on diaphragmatic motor function. We assessed the overall impact on the ability to respond to acute respiratory challenges (hypercapnia and hypoxia) using whole-body plethysmography.The chemogenetic stimulation of cervical eINs in a chronic cSCI model resulted in a significant improvement in ventilation. This demonstrates the feasibility of implementing the DREADD system to target spinal eINs under the CAMKIIa promoter in wild-type mice, indicating the translational potential of this approach. Our study presents a promising therapeutic strategy for enhancing respiratory function after SCI, utilizing a clinically relevant model of chronic cSCI. This represents a crucial step towards determining the future translation of this chemogenetic approach in individuals with chronic cSCI. The safety profile of the AAVs tested in this study, combined with the existence of FDA-approved AAV-based therapeutics, further supports their potential for clinical application.

Disclosures: K. Konkel: None. **A. Brezinski:** None. **S.N. Kurpad:** None. **K. Satkunendrarajah:** None.

Poster

PSTR137. Spinal Cord Injury—Therapeutic Strategies In Vivo: Pharmacological

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR137.04/K4

Topic: C.11. Spinal Cord Injury and Plasticity

Support: NIH Grant R01NS105961
NIH Grant 1R01NS122813
Shriners Research Foundation 85133-PHI-21
Wing For Life Spinal Cord Research Foundation WFL-US-09/21-249
Catalytic Collaborative Research grant at Temple University

Title: Treatments with novel β-catenin activators enhance axon regrowth and locomotor recovery in adult rodents with spinal cord injury

Authors: *H. KIM[1,2], H. NORISTANI[1,2], E. L. WALDEN[1,2], S. WANG[1,2], E. NURMEMMEDOV[3], S. LI[1,2];
[1]Dept. of Neural Sci., Temple Univ., Philadelphia, PA; [2]Shriners Hosp. Pediatric Res. Ctr., Philadelphia, PA; [3]Sigma Innotech LLC, Middletown, DE

Abstract: Patients with spinal cord injury (SCI) usually suffer from persistent functional deficits without a cure. This project is aimed at identifying effective drugs for promoting SCI recovery by targeting β-catenin, a dual-function protein that regulates cell-cell adhesion and gene transcription. β-catenin is widely expressed in many tissues, including CNS neural cells. It may

mediate neurogenesis, synaptic plasticity, and neurite outgrowth, but its overall roles in neural repair after CNS injuries are largely unknown. In this project, we use the novel selective β-catenin activators to evaluate whether β-catenin activation in the spinal cord promotes axon regrowth and locomotor functional recovery in adult mice with SCI. We performed dorsal over-hemisection at T7 in adult C57BL/6 mice and applied each of the two novel small beta-catenin activators (called BCA-18 and BCA-124) to the lesioned spinal cord locally. We have verified that these two compounds act cellularly by trapping the β-catenin transcriptional complex in activation mode. Six weeks after SCI, we evaluated the regrowth of multiple descending axons in the spinal cord caudal to the lesion. We found that treatment with β-catenin activators, especially BCA-18, significantly increased the numbers of serotonin axons and tyrosine hydroxylase axons in the transverse sections of the spinal cord 5-7 mm caudal to the lesion. BCA-18 treatment also moderately enhanced the regeneration of corticospinal tract axons 1-4 mm caudal to the lesion. Moreover, β-catenin activators significantly attenuated the sizes of reactive astrocytic scar tissues around the lesions. We evaluated functional recovery in a blinded manner and detected the enhanced locomotor BMS scores in the SCI mice treated with β-catenin activators. β-catenin activators also improved the functional recovery assessed by other behavioral tests, including increased grasping rates and reduced grid walk errors of the hindlimbs. Therefore, local treatments with selective β-catenin activators improve the regrowth of various motor axonal tracts and functional recovery in adult mice with SCI. Our findings indicate that β-catenin may become an attractive molecular therapeutic target for CNS injuries, including SCI.

Disclosures: H. Kim: None. **H. Noristani:** None. **E.L. Walden:** None. **S. Wang:** None. **E. Nurmemmedov:** None. **S. Li:** None.

Poster

PSTR137. Spinal Cord Injury—Therapeutic Strategies In Vivo: Pharmacological

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR137.05/K5

Topic: C.11. Spinal Cord Injury and Plasticity

Support: MN SCI & TBI OHE 143485

Title: Intranasal Insulin Improves Recovery in a Mouse Model of Cervical Spinal Cord Injury

Authors: J. M. FINE[1], T. BOWE[2], K. FALTESEK[2], E. CHRENKA[2], S. JACKSON[4], W. H. FREY, II[3], *L. HANSON[2];
[1]HealthPartners Neurosci. Ctr., [2]HealthPartners Inst., St. Paul, MN; [3]HealthPartners Neurosci., HealthPartners Inst., SAINT PAUL, MN; [4]Regions Hosp., St. Paul, MN

Abstract: A major obstacle to developing new treatments for spinal cord injury is the blood-brain barrier, which limits many potentially beneficial molecules from reaching the central nervous system. Intranasal administration of therapeutics is an innovative approach to rapidly and non-invasively bypass this barrier, and target drug delivery to the brain and spinal cord.

Insulin has anti-inflammatory and neuroprotective properties. Intranasal insulin has been shown to reach the cervical spinal cord at therapeutic concentrations after intranasal administration in mice. In this project we determined the effect of daily intranasal insulin treatment on recovery after cervical spinal cord injury (SCI) using a C7 impact model in female C57 mice. Behavior performance was evaluated 1 day prior to surgery and 1 week post-surgery with Basso mouse scale (BMS), inclined plane, and horizontal ladder. BMS was also assessed 24 hours post injury to confirm injury prior to initiation of daily treatment with intranasal insulin (INI; 2.4 IU or 0.6 IU) or saline (control) for one week. Tissue was collected for biochemical analysis. A group of surgical sham mice was included to evaluate the effects of the impact model. The SCI led to deficits in all three behavioral outcomes, with poorer performance in the SCI-Saline group as expected. After one week of treatment, two-way repeated measures ANOVA identified a significant change in BMS between all three SCI groups ($p = 0.03$), with the high dose insulin mice most improved. The same results were seen on the inclined plane test ($p = 0.02$). There were no significant differences in the horizontal ladder testing between the three SCI groups. A commercially available multiplex kit for 32 inflammatory markers was conducted on the injured portion of the spinal cord (1 mm), as well as the 1 mm both caudal and rostral to the injury. Most of the markers were modified in response to the injury, and the biggest changes in response to the insulin treatment included interleukin-9, a pro-inflammatory cytokine, and eotaxin, a chemokine ($p < 0.05$) as measured with ANOVA with Tukey's post-test. Overall, INI offers a promising route for the treatment of SCI.

Disclosures: **J.M. Fine:** None. **T. Bowe:** None. **K. Faltesek:** None. **E. Chrenka:** None. **S. Jackson:** None. **W.H. Frey:** None. **L. Hanson:** None.

Poster

PSTR137. Spinal Cord Injury—Therapeutic Strategies In Vivo: Pharmacological

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR137.06/K6

Topic: C.11. Spinal Cord Injury and Plasticity

Support: NINDS/NIH grant 5R01 NS111037

Title: Effect of repeat treatment of Rolipram loaded PgP nanoparticles via intrathecal adminstration on motor function and secondary injury in a female rat contusion SCI model

Authors: *Z. LIAO[1], C. JONES[1], B. ELLIOTT[1], K. HENRIE[1], M. R. DETLOFF[2], K. WEBB[1], J. LEE[1];
[1]Bioengineering, Clemson Univ., Greenville, SC; [2]Drexel Univ. Col. of Med., Drexel Univ. Col. of Med., Philadelphia, PA

Abstract: Spinal cord injury (SCI) disrupts axonal pathways, leading to permanent motor, sensory, and autonomic dysfunction. Several complex pathophysiological mechanisms included reduction of cyclic adenosine monophosphate (cAMP), neuroinflammation, and astrogliosis limit

spontaneous recovery. In our lab, we developed an amphiphilic polymeric nanocarrier, poly (lactide-co-glycolide)-graft-polyethylenimine (PgP) for delivery of the phosphodiesterase inhibitor, rolipram (Rm) to preserve/restore cAMP. While SCI is most common in males, the injury development, neurological recovery, and other crucial health outcomes are significantly vary depending upon sex. In this study, we evaluated effect of Rm-PgP single and repeat treatment via intrathecal catheter on neuroprotection and motor function recovery and neuropathic pain a in a female rat SCI model. A moderate T9 contusion model was generated by impacting at T9 spinal cord (200 kdyne, IH-0400 impactor, PSI). Intrathecal catheters (32 G, ReCath Co) were inserted at lumbar level (L4-5) through a hole made in the dura. Rats were divided into 4 groups: 1) sham, 2) untreated SCI (saline, 40 µl), 3) Rm-PgP-S: Rm-PgP (20 µg Rm, 40 µl) single injection immediately post-injury (time 0), 4) Rm-PgP-R: Rm-PgP (20 µg Rm, 40 µl) repeat injection at 0 and 1 week post-injury (WPI). Rm-PgP or saline was injected using microinjection pump with Hamilton syringe (28 G). Motor function and neuropathic pain were evaluated using Basso Bettie and Bresnahan (BBB) scoring system and von Frey test, respectively up to 6wks. At 6 WPI, rats were sacrificed via cardiac perfusion and the spinal cords retrieved for histological analysis. Another set of experimental groups was sacrificed by decapitation under deep anesthesia at 2 WPI and fresh spinal cords were harvested for measurement of cAMP level by ELISA. cAMP levels in both Rm-PgP-S and Rm-PgP-R treatment groups were higher than that in untreated SCI group. Both Rm-PgP treatment groups showed higher BBB scores than that in untreated SCI group. For the neuropathic pain, both Rm-PgP treatment groups showed lower pain levels than that in untreated SCI group. We observed that both Rm-PgP treatment groups showed increased spared myelin area, increased number of $NeuN^+$ cells and reduced fluorescence intensity from $GFAP^+$ activated astrocytes compared to untreated SCI. In conclusion, both Rm-PgP-S and Rm-PgP-R treatment via intrathecal administration increased average spared myelin area and neuronal cell survival, reduced astrogliosis, and improved motor function, and reduced neuropathic pain in a female rat moderate contusion SCI model.

Disclosures: **Z. Liao:** None. **C. Jones:** None. **B. Elliott:** None. **K. Henrie:** None. **M.R. Detloff:** None. **K. Webb:** None. **J. Lee:** None.

Poster

PSTR137. Spinal Cord Injury—Therapeutic Strategies In Vivo: Pharmacological

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR137.07/K7

Topic: C.11. Spinal Cord Injury and Plasticity

Support: State of Minnesota Office of Higher Education, Spinal Cord Injury and Traumatic Brain Injury Research Program

Title: Neurod1-mediated functional recovery after spinal cord injury

Authors: *A. ROMAN, A. PARR, A. GRANDE, W. LOW;
Univ. of Minnesota Twin Cities, Minneapolis, MN

Abstract: Spinal cord injury (SCI) remains a significant global concern with no available treatments capable of restoring nervous system function after injury. As a result, a major goal for SCI research is the regeneration of the spinal cord for functional recovery after injury. To address this, viral vector-mediated delivery of proneural factors for in vivo reprogramming has emerged as a potential experimental approach to promote restoration after central nervous system (CNS) injury. Current literature indicates that NeuroD1, a developmental proneural factor, is sufficient to convert astrocytes into neurons in vitro and in several animal models of CNS injuries and disorders. However, subsequent studies have highlighted an inconsistency in reprogramming efficiency and ambiguity in the cellular origin of "reprogrammed cells." In response, researchers have identified the viral dosage and the intervention timing as significant factors that influence reprogramming efficacy and reproducibility. We hypothesize that the AAV9-NeuroD1-based reprogramming platform is capable of reprogramming spinal cord astrocytes into neurons following SCI in a dose-dependent manner to restore nervous system function. Here we employed a two-vector, AAV9 DIO/FLEx-based delivery platform for selective expression of NeuroD1-mRuby2 (reprogramming) or mRuby2 alone (control) in spinal cord astrocytes after moderate SCI in female rats. Immunohistochemistry and microscopy-based analyses were used to examine viral transduction throughout the spinal cord at one of two viral titers (10^{13} or 10^{11} GC/mL) and timepoints (acute or subacute stage of injury). Despite the presence of successfully transduced cells in the gray matter of NeuroD1-treated spinal cords 6 weeks post-contusion injury, our preliminary motor functional analysis did not indicate any trend toward motor functional recovery. Future work will continue motor and sensory functional analysis and explore the spatial pattern of NeuroD1-mediated reprogramming through spatial transcriptomics and neuronal lineage tracing.

Disclosures: **A. Roman:** None. **A. Parr:** None. **A. Grande:** None. **W. Low:** None.

Poster

PSTR137. Spinal Cord Injury—Therapeutic Strategies In Vivo: Pharmacological

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR137.08/K8

Topic: C.11. Spinal Cord Injury and Plasticity

Support: Indiana Traumatic Spinal Cord & Brain Injury Research Grant Program, Grant Number 55051
Indiana University/Purdue University, Signature Center Initiative-Center for Spinal Cord and Brain Injury Research

Title: Differential Effects of Exercise and Hormone Treatment on Spinal Cord Injury-Induced Changes in Micturition and Morphology of External Urethral Sphincter Motoneurons

Authors: *E. HIBBARD[1], X. DU[2], Y.-H. ZHANG[2], X. M. XU[2], L. DENG[2], D. R. SENGELAUB[1];
[1]Indiana Univ., Bloomington, IN; [2]Indiana Univ. Sch. of Med., Indianapolis, IN

Abstract: Spinal cord injury (SCI) results in lesions that destroy tissue and spinal tracts, leading to deficits in locomotor and autonomic function. We have previously shown that after SCI, surviving motoneurons innervating hindlimb muscles exhibit extensive dendritic atrophy, which can be attenuated by treadmill training or treatment with gonadal hormones post-injury. We have also shown that following SCI, both exercise and treatment with gonadal hormones improve urinary function. Animals exercised with forced running wheel training show improved urinary function as measured by bladder cystometry and sphincter electromyography, and treatment with gonadal hormones improves voiding patterns as measured by metabolic cage testing. Here we tested if exercise or hormone treatment after contusive SCI results in similar protective effects on the structure and function of motoneurons innervating the external urethral sphincter (EUS). Gonadally intact young adult male rats received either a sham or a thoracic contusion injury. Immediately after injury, one cohort of animals was implanted with subcutaneous Silastic capsules filled with estradiol (E) and dihydrotestosterone (DHT) or left blank; continuous hormone treatment occurred for 4 weeks post-injury. This treatment regime was chosen as we have previously shown that combined treatment with E+DHT was maximally effective in protecting voiding patterns. A separate cohort of SCI-animals received either 12 weeks of forced wheel running exercise or no exercise treatment starting two weeks after injury. This regimen was chosen because improvement in nonlocomotory systems requires longer training than that required for locomotor improvement. At the end of treatment, urinary void volume was measured using metabolic cages and EUS motoneurons were labeled with cholera toxin-conjugated horseradish peroxidase, allowing for assessment of dendritic morphology in three dimensions. Void volumes increased after SCI in all animals, but were dramatically improved by treatment with E+DHT; in contrast, exercise was ineffective. Similar to what we have previously reported for hindlimb motoneurons after SCI, dendritic length of EUS motoneurons decreased by an average of 40% after SCI compared to sham animals, and this atrophy was prevented by treatment with E+DHT; in contrast, exercise was again ineffective. These results suggest that forced running wheel exercise is not a sufficient intervention to prevent SCI-induced changes in void volume or dendritic morphology in EUS motoneurons, whereas gonadal hormones could be used as an effective treatment for autonomic dysfunction after SCI.

Disclosures: E. Hibbard: None. **X. Du:** None. **Y. Zhang:** None. **X.M. Xu:** None. **L. Deng:** None. **D.R. Sengelaub:** None.

Poster

PSTR137. Spinal Cord Injury—Therapeutic Strategies In Vivo: Pharmacological

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR137.09/K9

Topic: C.11. Spinal Cord Injury and Plasticity

Support: NIH 1R01HL139708-01A1 (DDF)
SCIRTS Craig H. Neilsen Foundation (SR)
Rita Allen Foundation Scholars Program Fund, a component fund of the Community Foundation of New Jersey (AM)
NIH NIBIB Trailblazer award (R21 EB031249) (AM)
2022 Urology Care Foundation Research Scholar Award Program and the Indian American Urological Association Sakti Das, MD Awards (FA)

Title: Ampakines therapy improves voiding function and coordination following acute spinal cord injury

Authors: *F. ALOM[1], S. RANA[2], R. C. MARTINEZ[2], D. D. FULLER[3], A. D. MICKLE[4];
[1]Physiological Sci., [3]Univ. of Florida, [4]Univ. of Flordia, [2]Univ. of Florida, Gainesville, FL

Abstract: Traumatic spinal cord injury (SCI) often results in bladder dysfunction, leading to urological complications and reduced quality of life. The fundamental problem is uncoordinated voiding due to damage to sensory and motor circuits that control coordination between the bladder and external urethral sphincter (EUS) function. Recently, ampakines, an allosteric modulator of AMPA receptors, have been shown to improve diaphragm functions in SCI rats. In the bladder, AMPA receptors are present in both the sensory and descending efferent control circuits. Here we hypothesized that ampakines could acutely stimulate voiding function that has been impaired due to thoracic contusion SCI. Adult female Sprague Dawley rats received a unilateral contusion of the T9 spinal cord (n=10). Bladder function with EUS electromyography (EMG) was assessed five days post-SCI under urethane anesthesia and continuous saline infusion (0.1 ml/min). First, we recorded baseline cystometry in SCI and intact spinal rats (n=8). Following SCI, the voiding functions were remarkably affected, demonstrated by a high micturition threshold, prolonged inter-contraction interval, and increased voided volume during cystometry recording. Then we assessed the effects of vehicle (HPCD) and low impact ampakine (CX1739) over 30 minutes to investigate whether it could restore the voiding functions in SCI. Intravenous administration of HPCD showed no significant effects on voiding functions in SCI and spinally intact rats. However, intravenous delivery of CX1739 (5, 10, or 15 mg/kg), the micturition threshold, voided volume, and the interval between bladder contractions were dose-dependently, and significantly reduced to near spinal intact levels. Furthermore, ampakine could initiate coordinated voiding in non-voiding SCI rats (n=3). SCI data were compared to responses in intact spinal rats and baseline cystomtery. Our findings suggest that modulating AMPA receptor function using ampakines can rapidly improve voiding capability at sub-acute time points following contusion SCI. These results may provide a new and translatable method for therapeutic targeting of bladder dysfunction acutely after SCI.

Disclosures: F. Alom: None. **S. Rana:** None. **R.C. Martinez:** None. **D.D. Fuller:** None. **A.D. Mickle:** None.

Poster

PSTR137. Spinal Cord Injury—Therapeutic Strategies In Vivo: Pharmacological

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR137.10/K10

Topic: C.11. Spinal Cord Injury and Plasticity

Support: EU Horizon2020
Wyss Zurich CeNeReg project
Wings for Life
Swiss Paraplegic Foundation

Title: The NISCI clinical trial - first results of anti-Nogo-A antibody treatment in acute, cervical spinal cord injured patients

Authors: *M. SCHWAB[1], A. CURT[2], N. WEIDNER[3];
[1]Univ. of Zurich, Schlieren, Switzerland; [2]Balgrist Univ. Hosp., Zurich, Switzerland; [3]Univ. of Heidelberg, Heidelberg, Germany

Abstract: Antibodies which neutralize the neurite growth and plasticity inhibitory function of the CNS membrane protein Nogo-A were shown to enhance sprouting, regenerative fiber growth and recovery of lost functions in spinal cord injured (SCI) rats, mice and non-human primates. An investigator initiated, randomized, placebo-controlled phase II clinical trial ('NISCI') with a human anti-human-Nogo-A antibody was conducted from 2019 to 2022 in 13 spinal cord injury centers in Germany, Switzerland, Spain and the Czech Republic. The antibody was applied by 6 intrathecal injections separated by 5-day intervals into the lumbar CSF space (45 mg antibody/injection), starting within the first 2-4 weeks after acute traumatic cervical SCI randomized at a 2:1 ratio (verum versus placebo). At baseline (mean 10 days after injury), 128 patients ranging from complete (AIS A) to incomplete SCI (AIS B-D) were stratified into 11 impairment-specific cohorts according to the expected upper extremity motor score (UEMS) recovery at 6 months by unbiased recursive partitioning (Tanandini et al, 2014). Patients with injury severity predicting high UEMS recovery values (>43/50 motor score points) following standard rehabilitation were excluded due to expected ceiling effects. While the aimed treatment effect of ≥6 UEMS point recovery above standard rehabilitation over the total study group (n=114) was not met, the preplanned subgroup analysis shows higher levels of UEMS recovery in defined patient cohorts with distinct patterns of injury severity. Gains in lower extremity motor scores, quality of life measures, walking and autonomic functions are currently being evaluated. Assessments of CSF antibody titers as well as descriptors of cord damage (neurophysiology, proteomics and neuroimaging) as related to outcomes will yield a basis for further clinical trials aiming to enhance the biological repair responses of the spinal cord and CNS after injury.

Disclosures: **M. Schwab:** None. **A. Curt:** None. **N. Weidner:** None.

Poster

PSTR137. Spinal Cord Injury—Therapeutic Strategies In Vivo: Pharmacological

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR137.11/L1

Topic: C.11. Spinal Cord Injury and Plasticity

Support: FAPESP # 2021/02754-9
FAPESP # 2018/05006-0

Title: Neuroprotective effects of N, N-dimethyltryptamine after ventral root avulsion and reimplantation with a heterologous fibrin biopolymer

Authors: *P. A. CARO[1], E. HUERTAS[2], B. BARRAVIERA[3], R. S. FERREIRA JR.[3], A. L. OLIVEIRA[1];
[1]Mol. and Morphofunctional Biol., Univ. of Campinas - Lab. of Nerve Regeneration, Campinas, Brazil; [2]Inst. of Chem., Univ. of Campinas, Campinas, Brazil; [3]Ctr. for the Study of Venoms and Venomous Animals (CEVAP), São Paulo State Univ., Botucatu, Brazil

Abstract: Central Nervous System (CNS) / Peripheral Nervous system (PNI) interface injuries are an important clinical problem and recovery outcomes are generally poor. Ventral root avulsion (VRA) is an experimental model of proximal axonal injury at the CNS/PNI interface that induces death of the majority affected motoneurons (MNs), triggering a strong glial reaction as well as synaptic detachment. Combined therapies may be useful in such cases; therefore, we investigated neuroprotection elicited by the association of root reimplantation with aid of a heterologous fibrin sealant biopolymer, and the effect of the N, N-dimethyltryptamine (DMT). DMT was extracted from *Mimosa tenuiflora* roots and the compound was analyzed using mass spectrometry, infrared spectroscopy, Raman spectroscopy and nuclear magnetic resonance. To evaluate the neuroprotective effects of the DMT, we performed a L4-L6 rhizotomy in adult female rats. Lumbar spinal cord was collected at 14 days post-injury (dpi) for Nissl staining and immunofluorescence analyses. The experiments were approved by the Institutional Committee for Ethics in Animal Use (CEUA/IB/UNICAMP/Brazil, protocol number 5921-1/2021). This proximal axotomy resulted in significant death, up to 80% of spinal MNs after injury and showed markedly increased glial reactivity and synaptic detachment. DMT in different doses (1; 2.5 and 5 mg/kg) and DMT in combination with the MAO inhibitor pargyline reduced MN death 14 dpi, mainly in the doses of 1mg/kg and 2.5 mg/kg. Analysis of microglia morphology showed that DMT reduced type V and increased type I profiles, indicating an enhancement of surveying over activated microglial cells. The glial reactivity directly influenced synaptic covering over the MNs, which was increased after DMT treatment, mainly at the dose of 1mg/kg. Importantly, the pharmacological benefit of the DMT was further enhanced when associated with root reimplantation. Overall, our results demonstrated that the DMT exerted neuroprotection, immunomodulation and synapse preservation in a VRA model. Our data support that combinatorial approaches result in a better outcome after root avulsion.

Disclosures: P.A. Caro: None. **E. Huertas:** None. **B. Barraviera:** None. **R.S. Ferreira Jr.:** None. **A.L. Oliveira:** None.

Poster

PSTR137. Spinal Cord Injury—Therapeutic Strategies In Vivo: Pharmacological

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR137.12/L2

Topic: C.11. Spinal Cord Injury and Plasticity

Support: Woodnext Foundation

Title: Gut metabolite treatment to reduce metabolic dysfunction and improve functional recovery after spinal cord injury.

Authors: *A. J. DOUTHITT, C. G. GEOFFROY;
Dept. of Neurosci. and Exptl. Therapeut., Texas A&M Univ. Syst. Hlth. Sci. Ctr., Bryan, TX

Abstract: Spinal cord injury (SCI) is a traumatic injury associated with paralysis, sensory dysfunction, and bladder and bowel dysfunction. Trauma to the spinal cord and disruption of cellular signaling results in alteration of the metabolite expression profile, inflammation, astrogliosis, and impacts neuron survival and function. SCI also induces microbiome changes, which in turn induces pro-inflammatory changes. Microbiota are key regulators of immune-mediated inflammation. Antibiotic-induced dysbiosis increases SCI severity, and probiotic improves recovery after SCI. This gut-spinal cord communication is believed to result from changes in microbiota metabolites, however, the molecular basis for this communication is not fully characterized. Dysbiosis is paralleled by increased bacterial translocation to the blood which can increase the inflammatory response locally (spinal cord) and systemically, induce astrogliosis and impair neuron survival. Therefore, drugs with the potential of modifying the changes in the metabolite profile represent an underexplored strategy to enhance recovery; targeting inflammation, astrogliosis, and neuron survival. This highlights the need to better understand the changes in the gut metabolites post-SCI in order to develop effective therapeutic interventions. Recent studies have shown that microbiota metabolites are active molecular determinants of cellular functions in vivo. However, the full array of activities for most individual metabolites has not been established. In this study, tryptophan-derived gut metabolites were tested in an *in vivo* thoracic-8 level SCI mouse model to assess injury outcome. Subjects were treated with vehicle control or drug daily via oral gavage, followed by a transcardial perfusion 6-weeks post injury. Daily treatments promoted functional recovery and reduced the development of metabolic dysfunction as measured by behavioral and histological analysis, respectively. Therefore, supplementing naturally-derived gut metabolites post-SCI could be utilized as an effective strategy to reduce the secondary damage in SCI.

Disclosures: A.J. Douthitt: None. **C.G. Geoffroy:** None.

Poster

PSTR137. Spinal Cord Injury—Therapeutic Strategies In Vivo: Pharmacological

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR137.13/L3

Topic: C.11. Spinal Cord Injury and Plasticity

Support: DOD W81XWH-21-1-0777

Title: Dopamine D3 receptor activation of the spinal ejaculation generator facilitates sexual reflexes.

Authors: *T. H. ETTEY[1,2,3], R. RICE[1,2], L. M. COOLEN[1,2,3];
[2]Brain Hlth. Res. Institute, Dept. of Biol. Sci., [3]Sch. of Biomed. Sci., [1]Kent State Univ., Kent, OH

Abstract: Spinal cord injury has a devasting effect on ejaculatory function. Ejaculation is a complex reflex controlled by the spinal ejaculation generator (SEG) located in the lumbar spinal cord. In male rats, mice and humans, the SEG comprises a cluster of specialized neurons referred to as lumbar spinothalamic (LSt) cells, which integrate sensory inputs generated during sexual activity into a coordinated autonomic and motor response that ultimately culminates in ejaculation. In turn, the SEG is influenced by supraspinal inputs from brainstem and hypothalamus areas, including serotonin and dopamine inputs. The current study aims to test the role of dopamine on ejaculatory function by localization of dopamine receptors in the SEG and determining effects of dopamine agonists on ejaculation. Fluorescent *in situ* hybridization was used to test the expression of dopamine receptors in LSt cells. Spinal cord tissues of sham (n=6) or thoracic contusion-injured (SCI; n=5) male rats were hybridized using RNAscope™ probes specific for galanin as a marker for LSt cells together with probes for dopamine receptors 1,2 and 3 (Drd 1, Drd2, and Drd 3). Confocal microscopy and Fiji image-analyses showed that LSt cells co-expressed mRNA for Drd1 (37%), Drd2 (80%), or Drd3 (52%) with no effects of SCI. We previously demonstrated that D2/3 receptor agonist 7-OH-DPAT facilitates ejaculation in sham controls and restores ejaculatory reflex in chronically spinal injured rats. Here, we test the hypothesis that D3 receptors are the critical receptors by examining effects of the specific dopamine D3 agonist Pramipexole in control rats. Adult male Sprague Dawley rats received a complete spinal transection at T6-T7, and bulbocavernosus muscle (BCM) activities were recorded after systemic injections of saline (n=8), PPX (0.1 (n=8); 0.3 (n=9); or 1.0 (n=8) mg/kg) or PD 128907 (n=7; 0.1 mg/kg) and after a subsequent dorsal penile nerve (DPN) stimulation. BCM activities were analyzed for number of bursts, events and latency to first burst. Results showed that injections of PPX or PD 128907, but not saline, triggered increased BCM bursting activity. However, effects of PD 128907 group were significantly higher compared to PPX groups, suggesting that both D2 and D3 receptor activation triggers ejaculation. Current studies are examining effects of PPX in SCI male rats. Together, these findings suggest that dopamine receptor agonist can be considered for treatment options of ejaculatory dysfunction following SCI. Funded by DOD W81XWH-21-1-0777

Disclosures: **T.H. Ettey:** None. **R. Rice:** None. **L.M. Coolen:** None.

Poster

PSTR137. Spinal Cord Injury—Therapeutic Strategies In Vivo: Pharmacological

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR137.14/L4

Topic: C.11. Spinal Cord Injury and Plasticity

Support: The General Insurance Association of Japan a Grant-in-Aid for Scientifc Research [grant number JP17K10913, JP20H03558, JP21K16699]
The Japan Agency for Medical Research and Development [grant numbers JP20gm6210004, JP19am0101093, JP18ae0101047h]
The Kobayashi Foundation

Title: High-throughput assay to screen potential drugs identifies papaverine as neuroprotection drug for spinal cord injury via blood-spinal cord barrier protection

Authors: *Y. SUZUKI[1], K. KADOYA[1], A. SOTOME[1], A. SAKURABA[1], T. ENDO[1], T. ASANO[1], S. OTSUGURO[2], K. MAENAKA[3], S. NAKAGAWA[4], N. IWASAKI[1];
[1]Dept. of Orthopaedic Surgery, Fac. of Med. and Grad. Sch. of Med., [2]Ctr. for Res. and Educ. on Drug Discovery, Dept. of Med. Pharmacol., [3]Fac. of Pharmaceut. Sci., Hokkaido Univ., Sapporo, Japan; [4]Pharmaceut. Care and Hlth. Sciences, Fac. of Pharmaceut. Sci., Fukuoka Univ., Fukuoka, Japan

Abstract: INTRODUCTION: Blood-spinal cord barrier (BSCB) protection could reduce secondary damage and promote recovery after spinal cord injury (SCI). The purpose of this study is 1) to develop high-throughput screening assay (HTSA) for identifying drugs to protect BSCB function, and 2) to screen 3,200 FDA drugs and determine whether the identified drug has therapeutic effects on SCI. METHODS: Human brain endothelial cells (ECs) were incubated on 96 well plates with various concentrations of hydrogen peroxide (H_2O_2), and viability, cytotoxicity, and live/dead ratios were measured to determine the optimal condition for the HTSA. 3,200 existing drugs were screened using this newly developed HTSA, and one of the top hit drugs were subject to subsequent dose dependency and oxygen-glucose deprivation (OGD) tests. Then, in vitro BSCB co-culture model from rat brains with or without a candidate drug was stressed under the OGD condition and trans-endothelial electrical resistance (TEER) and Na-F permeability were measured. Next, C57BL/6 adult mice received intraperitoneal injections of a candidate drug or control, followed by SCI by wire-knife, and histological evaluation was performed the next day. Furthermore, rats received C5 contusion injury and 7 days of intraperitoneal injections of a candidate drug or control, followed by histological and functional analysis 2 and 8 weeks later. To investigate the molecular mechanism, RNA sequence of human brain ECs stimulated by the drug was performed. RESULTS: 450μM was determined as the optimal concentration of H_2O_2 for HTSA, based on the fact that its Z'-factor, S/B ratio, and CV were 0.75, 2.9, and 4% respectively. Papaverine, smooth muscle relaxant, was identified as the candidate drug in HTS, and protected human ECs from the OGD stress. Moreover, Papaverine maintained TEER and the Na-F permeability more than a control under OGD condition. Subjects treated with the drug demonstrated significantly smaller IgG leakage, glial scar, cystic cavity volume, better gait and pain behavior than control subjects after SCI. RNA sequence and in vitro verification results revealed that an unidentified drug action underlined the protection of the BSCB. DISCUSSION: Thus, the newly developed HTSA can successfully identify candidate drugs to protect human brain ECs from toxic stresses and identify Papaverine as a potential drug to protect BSCB functions from SCI. Though the drug is known as intracellular cAMP inducer, a

different mechanism underlies the protective effect on the BSCB. Since the drug is clinically applicable for acute SCI, it has a potential to be translated for SCI therapy.

Disclosures: Y. Suzuki: None. **K. Kadoya:** None. **A. Sotome:** None. **A. Sakuraba:** None. **T. Endo:** None. **T. Asano:** None. **S. Otsuguro:** None. **K. Maenaka:** None. **S. Nakagawa:** None. **N. Iwasaki:** None.

Poster

PSTR137. Spinal Cord Injury—Therapeutic Strategies In Vivo: Pharmacological

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR137.15/L5

Topic: C.11. Spinal Cord Injury and Plasticity

Support: Merit Review Award I01 BX002356
I01 BX003705
Mari Hulman George Endowment Funds (XMX)
Indiana Spinal Cord & Brain Injury Research Fund from ISDH (NKL)

Title: Mitochondria-targeted, cardiolipin-protecting peptide, SS-31, provides neuroprotection within in-vitro and in-vivo rodent models of spinal cord injury

Authors: *B. B. RAVENSCRAFT[1], D.-H. H. LEE[3], H. DAI[2], X.-M. M. XU[1], N. K. LIU[1]; [1]Neurolog. Surgery, [2]Indiana University-Purdue Univ. Indianapolis, Indianapolis, IN; [3]Baylor Col. of Med., Houston, TX

Abstract: Mitochondria-targeted peptide, SS-31, provides neuroprotection within in-vitro and in-vivo rodent models of spinal cord injury
Authors: B. RAVENSCRAFT, D.H. LEE, H. DAI, X.M. XU, N.K. LIU; Neurological Surgery, Indiana University School of Medicine, Indianapolis, IN
Disclosures: B. Ravenscraft: None. **D.H. Lee**: None. **H. Dai**: None. **X.M. Xu**: None. **N.K. Liu**: None.
Abstract: Traumatic spinal cord injury (SCI) is a severe medical problem with high mortality, long term morbidity, and locomotor deficits. Neuroprotective approaches have been increasingly emphasized for treating secondary cascades that follow the initial injury event. A promising neuroprotective approach is to target mitochondria, the effective gateways of apoptosis. Szeto-Schiller peptide 31 (SS-31) is a mitochondria-targeted peptide that has been shown to be beneficial in many pathology models, yet despite that exploration, the therapeutic has been relatively underexplored in SCI, with mixed results of basic locomotor functional recovery using doses limited to only 5mg/kg and early endpoints. It was unclear if a larger dose would further improve locomotion after SCI across acute to chronic injury timepoints.
For the first time, SS-31 was tested within primary spinal cord neuronal cultures, specifically from embryonic day 15 rat embryos. SS-31 provided protection against rotenone and glutamatergic excitotoxicity injury models, detected using methods to measure cell death

[Lactate Dehydrogenase (LDH) assay], cell viability [3-[4,5-dimethylthiazol-2-yl]-2,5 diphenyl tetrazolium bromide (MTT) metabolic assay], neuronal apoptosis [Caspase-3/-7 Glo assay], and neurite morphology [Neurotrack analysis]. SS-31 was then translated into an in-vivo mouse model of thoracic-level-10 (T-10) 60kdyne contusive SCI, where SS-31-mediated effects were measured at 1-day post-SCI with lipidomics, immunohistochemistry, and immunoblotting, while long-term effects were measured between 3-days post-SCI and 8-weeks post-SCI with several locomotor assays [Basso Mouse Scale, Grid Walk, Roto-Rod, & Tread-Scan], and histological analysis of lesion volume at 8-weeks post-SCI. In summary, immediate administration of SS-31 demonstrated neuroprotective effects within in-vitro and in-vivo models of rodent spinal cord injury, and notably the previously untested higher concentrations of SS-31 (100-200uM in-vitro and 10mg/kg in-vivo) provided the most robust neuroprotection.
Emphasizing the importance of the development of therapeutics that target mitochondrial functions, this study contributes insights into the beneficial effects of higher doses of SS-31.

Disclosures: B.B. Ravenscraft: None. **D.H. Lee:** None. **H. Dai:** None. **X.M. Xu:** None. **N.K. Liu:** None.

Poster

PSTR137. Spinal Cord Injury—Therapeutic Strategies In Vivo: Pharmacological

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR137.16/L6

Topic: C.11. Spinal Cord Injury and Plasticity

Support: HL146114

Title: Novel Regenerative Drug, SPG302 Promotes Functional Recovery of Diaphragm Muscle Activity After Cervical Spinal Cord Injury

Authors: *M. FOGARTY[1], W.-Z. ZHAN[2], V. F. SIMMON[3], P. VANDERKLISH[3], S. SARRAF[3], G. C. SIECK[2];
[2]Mayo Clin., [1]Mayo Clin., Rochester, MN; [3]Spinogenix, Inc, San Diego, CA

Abstract: Spinal cord hemisection at C_2 (C_2SH) is widely used to investigate the effects of reduced phrenic motor neuron (PhMN) activation on diaphragm muscle (DIAm) function, with reduction in DIAm activity on the injured side during eupnea. Following C_2SH, recovery of DIAm EMG activity may occur spontaneously over subsequent days/weeks. Various strategies have been effective at improving the incidence and magnitude of DIAm recovery during eupnea, but little is known about the effects of C_2SH on transdiaphragmatic pressure (P_{di}) during other ventilatory and non-ventilatory behaviors. We employ SPG302, a patent-protected novel small molecule, to assess whether enhancing synaptogenesis (i.e., enhancing spared local connections) will improve the incidence and the magnitude of recovery of DIAm EMG activity and P_{di} function 14-days post C_2SH. In anesthetized Sprague Dawley rats, DIAm EMG and P_{di} were assessed during eupnea, hypoxia/hypercapnia and airway occlusion prior to surgery (C_2SH or

sham [*n*=7]), immediately post-surgery and at 14-days post-surgery. In C_2SH rats, 14 days of DMSO (vehicle [*n*=14]) or SPG302 (*n*=10) treatments (IP injection) occurred. At the terminal experiment, maximum P_{di} was evoked by bilateral phrenic nerve stimulation. We show that significant EMG and P_{di} deficits during eupnea (~4% of initial EMG, 45% of initial P_{di}), HH (~5% of initial EMG, 45% of initial P_{di}) and occlusion (~20% of initial EMG, 45% of initial P_{di}) are apparent in C_2SH compared to sham rats immediately after surgery. In C_2SH rats treated with SPG302, recovery of eupneic, HH and occlusion DIAm EMG was enhanced ~2.5-fold compared to vehicle rats after 14 days. Treatment with SPG302 also ameliorated Pdi deficits following C_2SH, with ~36% improvement compared to vehicle treatment during eupnea, ~40% during HH, ~25% during occlusion and ~14% during maximum P_{di}. In summary, SPG302 is an exciting new therapeutic option for use to promote recovery after spinal cord injuries.

Disclosures: **M. Fogarty:** None. **W. Zhan:** None. **V.F. Simmon:** E. Ownership Interest (stock, stock options, royalty, receipt of intellectual property rights/patent holder, excluding diversified mutual funds); Spinogenix, Inc. **P. Vanderklish:** E. Ownership Interest (stock, stock options, royalty, receipt of intellectual property rights/patent holder, excluding diversified mutual funds); Spinogenix, Inc. **S. Sarraf:** E. Ownership Interest (stock, stock options, royalty, receipt of intellectual property rights/patent holder, excluding diversified mutual funds); Spinogenix, Inc. **G.C. Sieck:** None.

Poster

PSTR137. Spinal Cord Injury—Therapeutic Strategies In Vivo: Pharmacological

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR137.17/L7

Topic: C.11. Spinal Cord Injury and Plasticity

Support: NIH Grant NS124630-01
NIH Grant R01NS103481
NIH Grant R01NS111776
Wallace H Coulter Center
The Miami Project to Cure Paralysis
State of Florida

Title: Developing a kinase inhibitor drug to promote CNS axon regeneration and treat spinal cord injury

Authors: *H. ALI[1], A. BADILLO MARTINEZ[2], I. WILLIAMS[1], J. LEE[1], J. BIXBY[1], V. P. LEMMON[3];
[1]Univ. of Miami, Miami, FL; [2]Univ. of Miami Neurosci. Grad. Program, Miami, FL; [3]Miami Project to Cure Paralysis, Univ. Miami Miller Sch. Med., Miami, FL

Abstract: Damaged axons in the central nervous system (CNS) typically fail to regenerate, leading to irreversible loss of neuronal connectivity and associated functions. There are no

approved drugs for encouraging axon regrowth to promote CNS repair and functional recovery. Regeneration is suppressed by the lack of neuron-intrinsic regenerative capacity and by the neuron-extrinsic inhibitory environment of the injured CNS. To address this problem, we developed a therapeutic strategy that co-targets kinases in both extrinsic and intrinsic signaling pathways. We identified a kinase inhibitor (RO48) with advantageous polypharmacology (co-inhibits multiple intended targets and avoids detrimental off-targets). RO48 strongly promoted neurite outgrowth in cultured rodent primary neurons and in human iPSC-derived neurons. RO48 also promoted corticospinal tract (CST) sprouting and/or regeneration in multiple mouse models of SCI. Notably, these *in vivo* effects, which were accompanied with improvements in sensory and motor functions, were seen in several independent experimental series performed in distinct laboratories at different times. Hit-to-lead studies revealed a clear structure-activity relationship (SAR), underscored the importance of the favorable polypharmacology profile, and identified a lead candidate that is currently in preclinical development. Major progress has included substantial elaboration of the SAR landscape, improvement in permeability and cellular potency, and elimination of some liability off-targets. The program is currently focused on optimizing the pharmacokinetic and *in vivo* target-engagement profiles ahead of expanded efficacy testing in a rat contusion SCI model.

Disclosures: **H. Ali:** E. Ownership Interest (stock, stock options, royalty, receipt of intellectual property rights/patent holder, excluding diversified mutual funds); Dr. Ali is an inventor on an international patent application filed by the University of Miami on the compounds described in this study.. **A. Badillo Martinez:** None. **I. Williams:** None. **J. Lee:** None. **J. Bixby:** E. Ownership Interest (stock, stock options, royalty, receipt of intellectual property rights/patent holder, excluding diversified mutual funds); Dr. Bixby is an inventor on an international patent application filed by the University of Miami on the compounds described in this study. **V.P. Lemmon:** E. Ownership Interest (stock, stock options, royalty, receipt of intellectual property rights/patent holder, excluding diversified mutual funds); Dr. Lemmon is an inventor on an international patent application filed by the University of Miami on the compounds described in this study.

Poster

PSTR138. Spinal Cord Injury: Biological Repair Strategies

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR138.01

Topic: C.11. Spinal Cord Injury and Plasticity

Support: AMED under Grant JP23bm1233008 and JP23ym0126118

Title: Pretreatment with hepatocyte growth factor enhances human induced pluripotent stem cells transplantation-mediated functional recovery after spinal cord injury

Authors: *Y. SUEMATSU[1], N. NAGOSHI[1], Y. SAIJO[2], M. SHINOZAKI[2], M. NAKAMURA[1], H. OKANO[2];
[1]Keio Univ., Shinjuku-ku, Japan; [2]Keio Univ., Tokyo, Japan

Abstract: Introduction)Neural stem/progenitor cells derived from human induced pluripotent stem cells (hiPSC-NS/PC) transplantation is a novel therapeutic strategy for spinal cord injury (SCI). However, the degree of functional recovery reported to date is only modest. HGF is a potent growth factor that promotes tissue regeneration, and it has been reported to suppress secondary damage after SCI through angiogenic and anti-apoptotic effects. Therefore, the aim of this study was to combine HGF and hiPSC-NS/PC transplantation to further improve the efficacy of hiPSC-NS/PC transplantation therapy for subacute SCI. Methods) Severe contusive SCI was induced in RNU nude rats, and recombinant human HGF protein was administered continuously into the subarachnoid space with an osmotic mini-pump immediately after SCI for 2 weeks. Histological and comprehensive RNA-Seq gene expression analyses were conducted 2 or 7 days after SCI. To assess the effectiveness of combined therapy, subsequent to the HGF administration, hiPSC-NS/PC were transplanted into the injured spinal cord at 9 days post-injury. We compared outcomes among the four groups: Control (HGF(-), Transplantation(-)), HGF alone, Transplantation alone, and Combination groups. Motor functions were assessed by the Basso, Beattie, Bresnahan (BBB) score, Digigait analysis, kinematics, and electrophysiological analysis using motor evoked potentials (MEPs) were performed. Histological analyses were performed at 12 weeks after SCI. Results) HGF-treated tissues in the acute phase showed higher vascularization, neurogenesis (increased expression of neural stem cells and neurogenesis-related genes) and anti-inflammatory (decrease in TNF-α, IL-1β, and IL-6 by down-regulation of IKK/NF-kappaB signaling pathway) by RNA-seq analyses at 2 and 7 days after SCI. In addition, HGF-treated tissue at 84 days post-injury showed prominent preservation of the lesion site compared to the control group. Combination therapy of HGF and hiPSC-NS/PC transplantation promoted the survival rate of graft cells, remyelination, synaptic activity, and regeneration of intraspinal nerve fibers. The synaptic formation between host and graft neurons was observed by immunoelectron microscopy. Furthermore, BBB score and kinematic analysis showed significant recovery of locomotor function at 84 days post-injury, and the MEPs test also showed high action potentials, which indicated enhanced neurotransmission. Conclusion)The combination therapy of HGF and hiPSC-NS/PC transplantation is a novel and highly promising therapeutic strategy for the treatment of acute to subacute severe SCI.

Disclosures: **Y. Suematsu:** C. Other Research Support (receipt of drugs, supplies, equipment or other in-kind support); Drugs provided by Kringle Pharma, Inc.. **N. Nagoshi:** None. **Y. Saijo:** None. **M. Shinozaki:** None. **M. Nakamura:** None. **H. Okano:** None.

Poster

PSTR138. Spinal Cord Injury: Biological Repair Strategies

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR138.02/L8

Topic: C.11. Spinal Cord Injury and Plasticity

Support: ANID/FONDECYT 11190300

Title: Development of a Biocompatible Cellulose Nerve Scaffold morpho-functionalized with soy protein. Morphological and Schwann Cell Compatibility Analysis

Authors: *F. J. DIAS[1,2], B. GUTIERREZ[3], P. MARTINEZ-RODRÍGUEZ[2], M. GONZÁLEZ[4], J. ALARCÓN[5], K. GODOY[4], D. CURY[6];
[2]Oral Biol. Res. Ctr. - Dent. Sch., [3]Master Deegre Student in Dent. - Dent. Sch., [4]BIOREN, [5]Res. Ctr. in Dent. Sci. - Dent. Sch., [1]Univ. de La Frontera, Temuco, Chile; [6]Anat. and Cell Biol., Inst. of Biomed. Sci. - Univ. of Sao Paulo, Sao Paulo, Brazil

Abstract: Introduction: Peripheral nerve injuries have a negative impact on quality of life. The development of Nerve Guide Conduits (NGC) as biocompatible tubular scaffolds has been considered an important adjuvant treatment for these injuries. There is evidence that an NGC prepared with Cellulose functionalized with soy protein generates a biocompatible tubular scaffold with characteristics that could enhance nerve regeneration. **Objective:** To develop tubular nerve guides from Ethyl-Cellulose/Isolated Soy Protein and analyze its morphology and the biocompatibility of Schwann cells. **Material and Methods:** The NGC was prepared using the solvent evaporation/dip-coating technique using Ethyl-Cellulose/acetone and isolated soy protein. The three-dimensional NGC morphology was analyzed using a scanning electron microscope (SEM - SU3500-Hitachi) and the biocompatibility was analyzed by the viability of the Schwann cells (SCL 4.1 F7) and SEM analysis. **Results:** The SEM analysis shows in the internal polymeric matrix with appropriate mechanical characteristics, the scaffolds were able to suture and present uniformly distributed pores (>100 µm in diameter). The biocompatibility analysis revealed that Schwann cells were kept viable for 2 days, located in the internal region of the NGC lumen. **Conclusions.** The development of NGC using the solvent evaporation dip-coating technique with ethyl-cellulose functionalized with soy protein generates porous tubular scaffolds biocompatible with Schwann cells. **Further Steps:** Future studies will be carried out to analyze the physio-chemical characteristics of these NGC, enrich them with neurotrophic factors, and finally be able to implant them in animal models for morpho-functional studies.

Disclosures: F.J. Dias: None. **B. Gutierrez:** None. **P. Martinez-Rodríguez:** None. **M. González:** None. **J. Alarcón:** None. **K. Godoy:** None. **D. Cury:** None.

Poster

PSTR138. Spinal Cord Injury: Biological Repair Strategies

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR138.03/M1

Topic: C.11. Spinal Cord Injury and Plasticity

Support: NIH Grant R01 NS120877
NIH Grant R21 NS107946
NIH Grant T32 GM 65841

a MN State Grant for SCI and TBI Research
Regenerative Medicine Minnesota Grant

Title: Spatial Transcriptomic Analysis of Experimental Spinal Cord Injury to Understand Microenvironmental Changes after Targeting the Thrombin Receptor

Authors: *C. CHOI[1,2], H.-N. KIM[2], H. YOON[2,3], E. TRIPLET[2], I. A. SCARISBRICK[2,4,3]; [2]Dept. of Physical Med. and Rehabil., [3]Dept. of Physiol. and Biomed. Engin., [4]Ctr. for Regenerative Biotherapeutics, [1]Mayo Clin., Rochester, MN

Abstract: The thrombin receptor (Protease activated receptor 1, PAR1) is a G-protein coupled receptor that is activated by proteolytic cleavage to elicit intracellular signaling. Although thrombin activates PAR1 with high affinity and is frequently elevated in the context of neural injury, little is currently understood regarding the downstream outcomes or whether PAR1 can be targeted to limit injury and foster repair. In recent studies, we critically evaluated the roles of PAR1 as a regulator of the response of the adult spinal cord to traumatic injury. Genetic blockade of PAR1 resulted in improvements in sensorimotor coordination after spinal cord injury (SCI). These improvements were coupled to signs of improved neuron preservation with increases in presynaptic proteins, growth cone associated protein, and signs of myelin resiliency and regeneration. To gain molecular insight into the potential modulatory roles for PAR1 in SCI, in this study, we performed whole transcriptome analysis using RNA-Seq with a focus on the injury epicenter, as well as regions above and below. Experimental contusion-compression SCI was elicited by application of a modified aneurysm clip (FEJOTA™, 8g closing force) to the L1/L3 spinal cord of 12-wk old female wild type mice, or mice with genetic knockout of PAR1. At 30 d after injury, we analyzed transcriptional changes in each unique region of SCI and in the age and sex matched uninjured spinal cord. Ingenuity Pathway Analysis (IPA) of the differentially expressed genes identified unique canonical pathways differentially enriched by PAR1 knockout in each injury microenvironment. For example, pathways associated with axonal sprouting and plasticity including EIF2 Signaling (FDR 2.0E-20), mTOR Signaling (FDR 7.9E-13), and PTEN Signaling (FDR 5.8E-08) were specifically enriched in the region above the injury epicenter in PAR1 knockouts. By contrast, in region below the injury epicenter, energy and metabolism signaling, including oxidative phosphorylation (FDR 6.3E-25), mitochondrial dysfunction (FDR 7.9E-19), and sirtuin signaling Pathway (FDR 2.0E-08) gene sets were enriched. In the injury epicenter, gene sets associated with Leukocyte extravasation signaling (FDR 6.6E-02), Agranulocyte Adhesion and Diapedesis (FDR 2.0E-04), and MIF-mediated Glucocorticoid Regulation (FDR 6.6E-02) were enriched. These findings suggest that genetic knockout of PAR1 alters the microenvironment of the injured spinal cord in a region-specific manner. The molecular pathways identified here are being investigated for their essential roles in improving sensorimotor function in PAR1 knockout mice after experimental injury to the spinal cord.

Disclosures: C. Choi: None. **H. Kim:** None. **H. Yoon:** None. **E. Triplet:** None. **I.A. Scarisbrick:** None.

Poster

PSTR138. Spinal Cord Injury: Biological Repair Strategies

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR138.04/M2

Topic: C.11. Spinal Cord Injury and Plasticity

Support: CIHR

Title: Promoting the Differentiation of Neural Progenitor Cells into Oligodendrocytes through the Induction of Olig2 Expression: A Transcriptomic Study Using RNA-seq Analysis

Authors: *K. PIECZONKA[1], M. KHAZAEI[3], M. G. FEHLINGS[2];
[2]Univ. of Toronto, [1]Univ. of Toronto, Toronto, ON, Canada; [3]Dept. of Genet. and Develop., Krembil Res. Institute, Univ. Hlth. Network, Toronto, ON, Canada

Abstract: Spinal cord injury (SCI) is associated with the loss of oligodendrocytes, which play key roles in myelination and in modulating interactions between glia and neurons. Neural progenitor cells (NPCs) are a promising source of cells for SCI treatment, due to their ability to replace the lost oligodendrocytes, neurons and astrocytes. However, the differentiation of NPCs into oligodendrocytes is often inefficient, whereby the majority of the cells differentiate into astrocytes following transplantation. Therefore, we aimed to enhance oligodendrocyte differentiation by generating an inducible oligodendrogenic NPCs (ioNPCs) in which the extent of oligodendrocyte differentiation could be carefully regulated. Human ioNPCs were prepared by engineering NPCs to express Olig2 under the control of the conditional doxycycline-inducible tet-ON promoter, in which doxycycline administration regulates Olig2 expression. The cells were then treated with doxycycline for 3, 7 or 10 days. Next, the cells were characterized in vitro and in vivo using a combination of (1) qRT-PCR analysis, (2) immunostaining, and (3) bulk RNA sequencing. (1) qRT-PCR analysis revealed that the expression of several genes involved in oligodendroglial lineage determination, including OLIG1, OLIG2 and PDGFRA, progressively increased with longer doxycycline treatment timelines. (2) Immunostaining showed the ratio of O1+ oligodendrocytes was significantly higher in the ioNPCs (39.44 ± 16.5%) compared to NPCs (24.73 ± 6.5%). (3) Bulk RNA sequencing revealed that a total of 521 genes were differentially expressed between ioNPCs and NPCs. Oligodendroglial genes such as OLIG1, PDGFA, and MYRF, as well as neuronal and astrocyte genes such as TUBB3, MAP2 and S100B, were amongst the differentially expressed genes. Gene ontology analysis identified pathways corresponding to oligodendrocyte cell fate commitment and spinal cord oligodendrocyte fate specification amongst the differentially expressed genes. Furthermore, gene set enrichment analysis revealed an enrichment in the gene expression signatures of distinct subpopulations of oligodendroglial cells, including differentiation-committed oligodendrocyte precursors, newly formed oligodendrocytes, and myelin-forming oligodendrocytes. In conclusion, our study suggests that ioNPCs are a promising source of adjustable cells in which the extent of oligodendroglial biasing can be regulated, thus making them an optimal cell therapy for SCI.

Disclosures: K. Pieczonka: None. **M. Khazaei:** None. **M.G. Fehlings:** None.

Poster

PSTR138. Spinal Cord Injury: Biological Repair Strategies

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR138.05/M3

Topic: C.11. Spinal Cord Injury and Plasticity

Support: NS108189
R25NS117356

Title: In vivo visualization of PTEN knockdown with inducible promoters

Authors: *M. METCALFE[1], L. P. HALBERS[2], K. COLE[3], J. PRESCHER[3], A. LUPTAK[3], O. STEWARD[4];
[1]Univ. California Irvine, Irvine, CA; [2]Univ. of California Irvine, irvine, CA; [3]Univ. of California Irvine, Irvine, CA; [4]Univ. of California Irvine, Irvine CA, CA

Abstract: Deletion or shRNA-mediated knockdown of the gene phosphate and tensin homolog (PTEN) in cortical motoneurons (CMNs) enable regeneration accompanied by recovery of function after spinal cord injury. Previous approaches have focused on permanent PTEN knockout, which can lead to pathophysiologies. Accordingly, we are developing strategies to enable transient knockdown using inducible expression of short hairpin RNA against PTEN (shPTEN) via the TetOn system, where transgene expression is contingent on doxycycline (Dox) delivery. Our approach to document dynamics of regulated transgene expression uses bioluminescence imaging, where Dox in drinking water (2 mg/ml) activates transduction of both the transgene (shPTEN) and luciferase. When luciferase is expressed in cells, delivery of luciferin triggers production of photons which can be measured non-invasively in living animals over time to define onset and offset of luciferase expression. We examined the sensitivity of the In-vivo Imaging System (IVIS) for detecting AAV-retro-mediated luciferase expression in the brain after cervical spinal cord injections (C5) and Dox administration. We report results of 3 separate studies in which mice (n=15) received intra-spinal cord injections of AAVrg-TetON-shPTEN/luciferase. Delivery of Dox induces expression of both luciferase and shPTEN. 2d post-AAVrg injection, injections of luciferin triggered luminescence only in mice that received Dox, confirming activation of expression; delivery of luciferin continued to induce luminescence for 44d confirming persistence of expression. Immunostaining for PTEN 17d post Dox revealed PTEN knockdown in the cells of origin of the corticospinal tract in the cortex. Upon Dox removal, luminescence in response to luciferin gradually decreased until it was no longer detectable 30d post Dox removal. In a subgroup of mice injected with AAVrg-TetON-shPTEN (n=3) that underwent one cycle of ON/OFF Dox, Dox was reintroduced 90d post Dox removal to evaluate whether transgene expression could be reactivated. At 9d post Dox re-administration, luminescence was detected, confirming reactivation. This approach, coupled with *in vivo* luminescence imaging enables precise temporal control of transgene activity to optimize timing of therapeutic interventions while avoiding potential negative consequences associated with long-term PTEN knockdown.

Disclosures: **M. Metcalfe:** None. **L.P. Halbers:** None. **K. Cole:** None. **J. Prescher:** None. **A. Luptak:** None. **O. Steward:** E. Ownership Interest (stock, stock options, royalty, receipt of intellectual property rights/patent holder, excluding diversified mutual funds); OS is a co-founder and has economic interests in the company Axonis Inc, a biotechnology which is seeking to develop therapies for spinal cord injury and other neurological disorders.

Poster

PSTR138. Spinal Cord Injury: Biological Repair Strategies

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR138.06/M4

Topic: C.11. Spinal Cord Injury and Plasticity

Support: NINDS Grant R01NS079702
NINDS Grant R01NS110385

Title: Hepatocyte growth factor protects respiratory neural circuitry and preserves diaphragm function following cervical spinal cord injury

Authors: ***S. J. THOMAS**[1], B. GHOSH[2], Z. WANG[3], M. YANG[3], J. NONG[3], J. SEVERA[2], M. C. WRIGHT[4], Y. ZHONG[3], A. C. LEPORE[2];
[1]Neurosci., Thomas Jefferson Univ. Grad. Neurosci. Program, PHILADELPHIA, PA; [2]Neurosci., Thomas Jefferson Univ., Philadelphia, PA; [3]Sch. of Biomed. Engineering, Sci. and Hlth. Systems, Drexel Univ., Philadelphia, PA; [4]Biol., Arcadia Univ., Philadelphia, PA

Abstract: A major portion of spinal cord injury (SCI) cases occur in the cervical region where essential components of respiratory neural circuitry are located. Phrenic motor neurons (PhMNs) housed in the C3-C5 spinal cord directly innervate the diaphragm, and SCI-induced damage to these cells severely impairs respiratory function. In this study, we tested a biomaterial-based approach towards preserving this critical phrenic motor circuitry after cervical SCI by locally delivering hepatocyte growth factor (HGF). HGF been found to possess a range of therapeutic capabilities relevant to nervous system repair, including anti-inflammatory, anti-fibrotic, and anti-apoptotic effects. We developed a hydrogel-based HGF delivery system that can be injected into the intrathecal space for sustained, local delivery of high levels of HGF without damaging the spinal cord. Implantation of HGF hydrogel after unilateral C5 contusion-type SCI in rats preserved both diaphragm function (as assessed by *in vivo* recordings of compound muscle action potentials and inspiratory electromyography amplitudes) and PhMN innervation of the diaphragm (as assessed by detailed neuromuscular junction morphological analysis and retrograde PhMN tracing). Furthermore, HGF hydrogel significantly decreased lesion size and degeneration of cervical motor neuron cell bodies, as well as reduced the levels of two scar-associated molecules surrounding the injury site: chondroitin sulfate proteoglycan, an inhibitor of axon growth capacity; and collagen type III, a marker of fibrotic scar formation. Our findings demonstrate that local biomaterial-based delivery of HGF hydrogel to the injured cervical spinal cord is a robustly effective strategy for preserving respiratory circuitry and diaphragm function.

Disclosures: **S.J. Thomas:** None. **B. Ghosh:** None. **Z. Wang:** None. **M. Yang:** None. **J. Nong:** None. **J. Severa:** None. **M.C. Wright:** None. **Y. Zhong:** None. **A.C. Lepore:** None.

Poster

PSTR138. Spinal Cord Injury: Biological Repair Strategies

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR138.07/M5

Topic: C.11. Spinal Cord Injury and Plasticity

Support: Fondo de Investigación Anahuac 201706

Title: Influence of symbiotic administration on gut microbiota composition and butyrate concentrations after spinal cord injury in different levels and intensities

Authors: *E. GARCIA-VENCES, E. DE LA CRUZ CASTILLO, R. H. RODRIGUEZ-BARRERA, A. FLORES ROMERO, A. IBARRA;
Facultad de Ciencias de la Salud, Univ. Anahuac Mexico Norte, Mexico city, Mexico

Abstract: Spinal cord injury (SCI) causes denervation of the autonomic nervous system (ANS) connection, leading to various complications such as changes in the gut microbiota. Gut dysbiosis and its restoration depend on the level and intensity of spinal cord injury, impacting on butyrate concentrations, which has been shown to have a neuroprotective effect in some models of cognitive impairment and neurodegenerative processes. Currently, the use of symbiotics to support gut microbiota balance and contribute to butyrate production has been shown to have different effects on neuroimmune pathways, neuroendocrine pathways, and microbiota-derived neurotransmitters allowing beneficial effects in neurodegenerative models. The aim of the present project was to characterize changes in gut microbiota and butyrate concentrations at different levels (T5 / T9) and injury intensities (moderate M and severe S) in rats after SCI, with a symbiotic treatment. Sprague Dawley rats were randomized into 5 groups Sham, T5M, T5S, T9M, and T9S. These were assessed using the BBB scale for locomotor recovery on a weekly until the end of the study. Four weeks after injury, fecal samples were collected and the animals received the symbiotics daily to eight weeks post-injury, collected fecal samples at this time. Butyrate concentrations were determined by gas chromatography and the gut microbiota was characterized by the DNA sequence of the 16s gene (regions 3 and 4). Regarding motor recovery at week four, was only a significant improvement in the T9M group compared to T5S; however, after supplementation at the end of the study, motor recovery of T9M had the greatest recovery, followed by T9S and T5M with no significant change for the T5S group. All experimental groups presented alterations in the relative abundance. Clostridium at four weeks decreased in all experimental groups with respect to Sham. At eight weeks the symbiotics increased the population of Bifidobacterium, Lactobacillus, and Clostridium; this improvement is not found in the same way in the T5S group. Regarding butyrate concentrations not showed differences between the groups; however, at eight weeks, butyrate levels were higher in both T9 groups, with a smaller increase in the T5 groups.

Disclosures: **E. Garcia-Vences:** None. **E. De la Cruz Castillo:** None. **R.H. Rodriguez-Barrera:** None. **A. Flores Romero:** None. **A. Ibarra:** None.

Poster

PSTR138. Spinal Cord Injury: Biological Repair Strategies

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR138.08/M7

Topic: C.11. Spinal Cord Injury and Plasticity

Title: Neurogenesis induced by immunization with peptide A91 in rats with spinal cord injury

Authors: *R. RODRIGUEZ BARRERA[1], S. SANCHEZ-NORIEGA[2], E. GARCIA-VENCES[2], Y. CRUZ MARTINEZ[2], M. GONZÁLEZ PACHECO[2], S. FLORES LÓPEZ[2], A. IBARRA[3];
[1]Univ. Anahuac Mexico, Mexico City, Mexico; [2]Univ. Anahuac Mexico Norte, Mexico city, Mexico; [3]Coordinación de Investigación, Facultad de Ciencias de la Salud, Univ. Anahuac-Centro de Investigación Camina, Estado De México, Mexico

Abstract: The objective of this study was to report the induction of neurogenesis through the use of modified neural peptide (INDP) A91, in spinal cord injuries (SCI) during chronic stages at the cervical, thoracic, and lumbar levels. Immunization with modified neural peptides has shown a promising therapy to achieve a regenerative effect on spinal cord injury. The Sprague Dawley strain female rats were used to cause them an injury by moderate contusion and subsequent immunization with peptide A91 (150 micrograms were applied on the tail base), and the motor function was tested using the BBB scale. Neurogenesis was analyzed with immunofluorescence, double labeling with anti-5 bromo-2'-deoxyuridine (BrdU), and doublecortin antibodies (Dcx). INDP induced significant production of anti-inflammatory and regeneration-associated proteins in the chronic stages of SCI at the cervical, thoracic, and lumbar levels. A trend towards improvement was observed from the second week in both groups, the group immunized with A91 had a better score statistically significant after four weeks of the SCI: [4.5 ± 1.3 mean and standard deviation error (SDE)], versus the group treated with PBS (3.5 ± 0.9 mean and SDE). Furthermore, during the neurogenesis evaluation with BrdU+/DCX+ double labeling in the cervical, thoracic, and lumbar spinal cord, active neuroblast formation was found even in those PBS-treated rats. Therefore, a higher number of them has been observed in rats immunized with A91. Nevertheless, both groups showed an increase, the statistically significant groups were: A91 thoracic level compared to PBS cervical level at 30 days after SCI, A91 thoracic level compared to PBS level thoracic at 30 days post-SCI and A91 thoracic level compared to PBS lumbar level at 30 days post-SCI. The induction of basal neurogenesis could indicate cells are forming neurons by traveling from higher centers such as the ZSV and the dentate gyrus of the hippocampus; these regions could be therapeutic targets to induce cell repair mechanisms in the CNS. An increase in lumbar regions would indicate that neurogenesis could be neurogenic niches in the spinal cord. This could help to improve existing therapies and it is suggested as a specific treatment in chronic phases.

Disclosures: **R. Rodriguez Barrera:** None. **S. Sanchez-Noriega:** None. **E. Garcia-Vences:** None. **Y. Cruz Martinez:** None. **M. González Pacheco:** None. **S. Flores López:** None. **A. Ibarra:** None.

Poster

PSTR138. Spinal Cord Injury: Biological Repair Strategies

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR138.09/M8

Topic: C.11. Spinal Cord Injury and Plasticity

Support: The Wings for Life Foundation under contract number WFL-US-13/22
The Craig H. Neilsen Foundation under award #LOIID 998439, the National Institute of Neurological Disorders and Stroke (NINDS) under Award: R01NS116068
The Spinal Cord and Brain Injury Research Center Endowed Chair #5

Title: Tissue clearing reveals the spatial distribution of axon sprouting after using retrogradely transported AAVs to knockout PTEN in a lateral hemisection model of spinal cord injury.

Authors: *C. C. BOSSE-JOSEPH, K. A. PARK, J. C. GENSEL, A. N. STEWART;
Spinal Cord & Brain Injury Res. Ctr., Univ. of Kentucky, Lexington, KY

Abstract: Spinal cord injury poses multiple regeneration barriers, including neuronal-intrinsic and extrinsic factors. Overcoming these barriers has been a longstanding challenge in neuroscience. One well-studied mechanism to promote spinal cord regeneration involves activation through the Akt/mTorc1 pathway by knocking out phosphatase and tensin homolog protein (PTEN). PTEN knockout (KO) via adeno-associated virus (AAV) viral vector has shown promising results, although further investigations are required. In this study, we utilized a mouse model of T9 hemisection and administered cre-recombinase (Cre) and a red fluorescent protein via AAV-retrograde (AAVrg) to induce PTEN knockout. Our primary objectives were three-fold: 1) Determine and compare the extent to which PTEN-KO using AAVrg's affects locomotor function in both the ipsilateral and contralateral leg after lateral hemisection. 2) Measure the extent of midline crossing of spared axons after PTEN-KO throughout the spinal axis below the lesion. 3) Directly compare the extent that AAVrg's can transduce spared versus damaged axons in spinal-projecting neurons throughout the brain and brainstem. To assess the behavioral recovery of the mice, we utilized weekly Basso, Beattie, and Bresnahan (BBB) locomotor rating scale scoring to evaluate the effectiveness of PTEN-KO in promoting functional recovery. Moreover, we optimized tissue-clearing techniques and employed confocal microscopy to visualize and reconstruct three-dimensional images of the brain and spinal cord, aiming to compare axon sprouting and transduced neuronal populations quantitatively. Our results shed light on the potential for AAVrg's to induce genetic knockouts in both damaged and spared axons and provide valuable insights into the role of cross-hemisphere sprouting in improving locomotor functions after PTEN-KO. By elucidating the extent of behavioral recovery and

assessing the spatial distribution of newly generated axons within the cord, this study contributes to the understanding of spinal cord regeneration and offers directions for developing novel therapeutic approaches. The results obtained will advance our knowledge of the utility of viral vectors for genetic manipulation in spinal cord injury.

Disclosures: C.C. Bosse-Joseph: None. **K.A. Park:** None. **J.C. Gensel:** None. **A.N. Stewart:** None.

Poster

PSTR138. Spinal Cord Injury: Biological Repair Strategies

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR138.10/M9

Topic: C.11. Spinal Cord Injury and Plasticity

Support: TARCC2020
NIH NS099073
NIH NS092616
NIH NS111776
NIH NS117065
NIH NS088095
NIH NS103481
ISDH58180

Title: Ng2 glia reprogramming induces robust axonal regeneration after spinal cord injury

Authors: *W. WU[1], X. DU[2], W. TAI[4], C. CHEN[3], C.-L. ZHANG[4], X. M. XU[3];
[1]Indiana Univ. Sch. of medicine, Indianapolis, IN; [2]Neurolog. Surgery, [3]Indiana Univ. Sch. of Med., Indianapolis, IN; [4]UT Southwestern Med. Ctr., UT Southwestern Med. Ctr., Dallas, TX

Abstract: Spinal cord injury (SCI) often leads to neuronal loss, axonal degeneration and behavioral dysfunction. We recently show that in vivo reprogramming of NG2 glia produces new neurons, reduces glial scaring, and ultimately leads to improved function after SCI. By examining endogenous neurons, we here unexpectedly uncover that NG2 glia reprogramming also induces robust axonal regeneration of the corticospinal tract and serotonergic neurons. Such reprogramming-induced axonal regeneration may contribute to the reconstruction of neural networks essential for behavioral recovery.

Disclosures: W. Wu: None. **X. Du:** None. **W. Tai:** None. **C. Chen:** None. **C. Zhang:** None. **X.M. Xu:** None.

Poster

PSTR138. Spinal Cord Injury: Biological Repair Strategies

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR138.11/M10

Topic: C.11. Spinal Cord Injury and Plasticity

Support: NIH R01-NS042291
NIH R01-NS104442
NIH NCRR P51 OD011107-56
DOD CDMRP SC170233
DARPA
Dr. Miriam and Sheldon G. Adelson Medical Research Foundation
Bernard and Anne Spitzer Charitable Trust
IP50RX001045
RR&D B7332R

Title: Grafts of Human Spinal Cord Neural Stem Cells into Primate Hemisection Spinal Cord Injury Improve Functional Outcomes After 3 Months

Authors: *E. SINOPOULOU[1], E. ROSENZWEIG[2], J. BROCK[2], P. P. LU[4], R. HUIE[5], A. TORRES-ESPIN[5], N. KYRITSIS[5], Y. NOUT-LOMAS[7], A. FERGUSON[5], M. BEATTIE[5], J. BRESNAHAN[6], M. TUSZYNSKI[3];
[1]UCSD, La Jolla, CA; [2]UCSD, san diego, CA; [3]UCSD, San Diego, CA; [4]UCSD Dept. of Neurosciences, La Jolla, CA; [5]UCSF, San fransisco, CA; [6]UCSF, San Fransisco, CA; [7]Colorado State Univ., fort collins, CO

Abstract: We have been developing a neural stem cell treatment approach for spinal cord injury (SCI) in which early stage neural stem cells (NSCs) are grafted into spinal cord lesion sites to form new neural relays across the lesion: transected host axons regenerate into a neural stem cell graft implanted into the lesion site, and form synapses; grafted neurons in turn extend axons out from the lesion site and into the distal host spinal cord and synapse onto host neurons. This circuitry establishes a basis for poly-synaptic relays across the lesion (Lu et al., *Cell* 2012; Lu et al., *Neuron* 2014; Kadoya et al., *Nat Med* 2016; Rosenzweig et al., *Nat Med* 2018). Key to this approach is the creation of a human neural stem cell line of spinal cord identity, **H9-scNSCs**, derived from the H9 embryonic stem cell line (Kumamaru et al, *Nat Meth* 2018). In the present study, we grafted this lead candidate human neural stem cell line into adult rhesus monkeys that underwent unilateral C7 spinal cord transection lesions. We then evaluated functional and anatomical outcomes after injury. N=10 monkeys underwent right C7 hemisection lesions as described (Rosenzweig et al, *Nat Med* 2010). Two weeks later, 4 monkeys received **H9-scNSC** grafts into the lesion site, and N=6 were lesioned controls. Over the next 150 days, grafted monkeys exhibited a significant, 2.5-fold improvement in retrieval of food objects on a Brinkman board using finger flexion compared to lesion controls ($P<0.01$, Beta mixed spline regression analysis). Anatomically, grafted subjects exhibited extension of up to hundreds of thousands of axons caudal to the lesion site for distances of up to 50 mm. Host axons labeled for neurofilament penetrated the grafts and formed synapses. Thus, anatomical conditions for neural relay formation were established. These findings support the efficacy of H9-scNSC grafts for SCI in primates. This program is undergoing clinical translation and a Pre-IND meeting with the FDA has been conducted.

Disclosures: **E. Sinopoulou:** None. **E. Rosenzweig:** None. **J. Brock:** None. **P.P. Lu:** None. **R. Huie:** None. **A. Torres-Espin:** None. **N. Kyritsis:** None. **Y. Nout-Lomas:** None. **A. Ferguson:** None. **M. Beattie:** None. **J. Bresnahan:** None. **M. Tuszynski:** None.

Poster

PSTR138. Spinal Cord Injury: Biological Repair Strategies

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR138.12/N1

Topic: C.11. Spinal Cord Injury and Plasticity

Support: NIH Grant 1R21NS116550

Title: Tissue-engineered repair with motor cortex-spinal cord dual neuromodulation promotes corticospinal system axon outgrowth in rats after cervical contusion SCI

Authors: *P. T. J. A. WILLIAMS[1], E. SCHELBAUM[1], H. ALEXANDER[1], K. KANTE[2], A. CARTER[1], H. SHARIF[1], S. SOARES[2], F. NOTHIAS[2], J. H. MARTIN[1];
[1]Cellular, Mol. and Biomed. Sci., CUNY Sch. of Med., New York, NY; [2]Neurosci. Paris Seine, CNRS-UMR8246, INSERM1130, Sorbonne Univ., NPS/IBPS, Paris Cedex 05, France

Abstract: Trauma caused by spinal cord injury (SCI) damages axons and their targets at the injury site. Most injuries are incomplete but the lesioned environment limits support for repair and growth, which leads to impairments chronically. The corticospinal tract (CST) is particularly vulnerable, and SCI weakens connections with circuits at and below the injury site. Our approach to repair the CS is twofold: to transform the trauma site into a more favorable environment that supports repair using biomaterial tissue-engineering, and to boost corticospinal tract outgrowth to deprived circuits using targeted neuromodulation. We injected an engineered chitosan-based hydrogel into the lesion site 3 days after moderate midline C4 contusion (200 kdyne) in rats. We then administered neuromodulation therapy - combining phasic patterned stimulation of the motor cortex (epidural intermittent theta burst; iTBS) concurrently with cathodal direct current stimulation of the spinal cord (transcutaneous spinal direct current stimulation; tsDCS) - during daily sessions (28 minutes) for 10 consecutive days starting the day after the biomaterial injection. We anterogradely labeled CST axons with viral tracers (AAV2 with CamKII promoter) and quantified their density and axon complexity in the chronic phase (8 weeks).There was remarkable repair of the lesion site with less scar formation and reduced cavitation, often near complete elimination, in the groups that received biomaterial injection. Caudal to the injury, we found increased axon length and branching in rats that received combined biomaterial injection with neuromodulation compared to each application alone. Ongoing analyses are examining if the lesion site repair also reduced CST axon dieback. In summary, transforming the lesion site with tissue engineering in combination with targeted neuromodulation to promote axon sprouting from spared tracts is a promising approach to achieve durable rewiring after SCI.

Disclosures: **P.T.J.A. Williams:** None. **E. Schelbaum:** None. **H. Alexander:** None. **K. Kante:** A. Employment/Salary (full or part-time); MedJeduse. **A. Carter:** None. **H. Sharif:** None. **S. Soares:** E. Ownership Interest (stock, stock options, royalty, receipt of intellectual property rights/patent holder, excluding diversified mutual funds); CNRS. **F. Nothias:** E. Ownership Interest (stock, stock options, royalty, receipt of intellectual property rights/patent holder, excluding diversified mutual funds); CNRS. **J.H. Martin:** None.

Poster

PSTR138. Spinal Cord Injury: Biological Repair Strategies

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR138.13/N2

Topic: C.11. Spinal Cord Injury and Plasticity

Support: NINDS, NIH; R01 NS104291 (MAL)
F31 NS125975 (TF)
F32 NS119348 (LVZ)
T32 NS121768 (AH)

Title: Initial donor-host connectivity of fetal tissue transplants after a cervical spinal cord injury is lost following an extended survival timeline.

Authors: *A. A. HALL[1], K. LOCKE[1], T. FORTINO[1], A. NICEFORO[1], K. SCHARDIEN[1], L. ZHOLUDEVA[1,2], M. LANE[1];
[1]Neurobio. and Anat., Drexel University, Col. of Med., Philadelphia, PA; [2]Gladstone Inst., San Francisco, CA, CA

Abstract: Cervical spinal cord injury disrupts motor, sensory, and autonomic networks leading to life-threatening deficits, such as impaired breathing which is the result of compromised phrenic (diaphragm) motor networks. Spontaneous plasticity/recovery has been observed but the extent of recovery remains limited. While therapeutic interventions such as activity-based therapy and neural stimulation can enhance this plasticity, they are reliant on spared neural tissues, leaving the underlying damage unaddressed. To address this, transplantation of developing neural cells to repair the injured CNS have been used to promote tissue repair, further promoting recovery. This strategy provides neural building blocks capable of novel pathway generation and reinnervation of disconnected regions caudal to the injury. The central hypothesis to our ongoing research is that transplanting neural progenitor cells enriched with spinal interneurons provides the necessary components for neural repair. The formation of donor relays, however, is reliant on stable synaptic integration between donor and host cells.. Though chronic transplantation studies have shown some efficacy, evidence for lasting donor-host connectivity remains limited. The goal of the present work is to address this gap in knowledge. Spinal interneuron-rich spinal neural progenitor tissue was transplanted into the injured cervical spinal cord to restore connectivity and function to the injured phrenic network. Spinal cord tissue dissected from E13.5 GFP-Sprague-Dawley embryos was transplanted into the lesion epicenter

1-week following a moderate lateral cervical contusion in adult Sprague Dawley rats. 1- or 12-months post-transplant, transneuronal retrograde viral tracing of the diaphragm with pseudorabies virus was performed to map the synaptic integration between donor neurons and the phrenic network. Preliminary results show that while donor cell survival was comparable between 1 and 12 months after delivery, there was an attenuation of donor-host connectivity to the phrenic system over time. Functional testing found no difference in diaphragm electromyographic (dEMG) output between year-long transplanted and control groups ipsilateral to injury. These findings suggest that transplanting cells alone is insufficient for lasting integration between transplanted neurons and injured spinal networks. Ongoing studies are exploring these changes in connectivity and assessing whether combining transplantation with activity-based therapies can stimulate greater, consistent, and persistent donor-host connectivity.

Disclosures: **A.A. Hall:** None. **K. Locke:** None. **T. Fortino:** None. **A. Niceforo:** None. **K. Schardien:** None. **L. Zholudeva:** None. **M. Lane:** None.

Poster

PSTR138. Spinal Cord Injury: Biological Repair Strategies

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR138.14/N3

Topic: C.11. Spinal Cord Injury and Plasticity

Support: PA-SCI

Title: Regenerative Capabilities of Virally Administered Retrograde CaRheb combined with Optogenetics in a complete Spinal Cord Injury Model

Authors: *J. D. PASTORINO, V. TOM, S. F. GISZTER;
Neurobio. and Anat., Drexel Univ. Col. of Med. Neurosci. Program, Philadelphia, PA

Abstract: Spinal cord injury(SCI) results in loss of sensation and function with no known complete cure. In order to promote spinal cord repair after injury, we look to induce neural growth from the motor cortex, provide a suitable bridge for this neuronal growth, bypass the CSPG barrier, stimulate the motor cortex, and restore functional hind limb activity. Optogenetic stimulation of motor cortex neurons may help engage cortex and restore impaired signaling to/from trunk/ hind limb circuits after the SCI. Viral constitutively active ras homolog (caRheb) injections specifically promote neuronal growth rostral to injury and combined with chondroitinase ABC (chABC) intrathecally delivered support growth beyond a peripheral nerve graft of pro-reparative Schwann cells. Retrograde AAV (rAAVretro) delivery of caRheb may target descending corticospinal and other neurons projecting to thoracic spinal cord. Activating lumbar central pattern generators (CPGs) via brain derived neurotrophic factor (BDNF) promotes stepping combined with robot rehabilitation therapy for strengthening step CPG circuitry and trunk muscle controls. Preliminary data shows no significant improvement over time in kinematic scores of animals who receive rAAVretro_caRheb without robot treadmill

training (n=4). After combining robot treadmill training with rAAVretro_caRheb treatment, kinematic scores are significantly higher in the last days of training compared to first (n=7). Animals that receive both rAAVrertro_caRheb, optogenetic subthreshold stimulation, and 6 weeks robot treadmill training significantly improve stepping on last day compared to first (n=4). Final scores of this group are currently trending higher than animals receiving only rAAVretro_caRheb and robot treadmill training. Kinematic scores also indicate treadmill training and the neural bridging interact with the other therapies, avoiding a late collapse in function as often seen BDNF without bridging. Imaging in the motor cortex reveals that there is consistent co-expression of rAAVretro_caRheb and mCherry/channelrhodopsin in cortex within all animals completed (n=30), allowing for analysis of soma size morphology changes between groups. Neural bridging to date replicates results of Dr. Houle and Co-PI Dr. Tom (Spinal Cord Research Center).

Disclosures: **J.D. Pastorino:** None. **V. Tom:** None. **S.F. Giszter:** None.

Poster

PSTR138. Spinal Cord Injury: Biological Repair Strategies

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR138.15/N4

Topic: C.11. Spinal Cord Injury and Plasticity

Support: R01NS115963
R01NS100772

Title: Restoration of Motor Function following Spinal Cord Injury through Harnessing Opsins with Ultra-Potent Light Sensitivity and Wireless Spinal Interface.

Authors: *A. AMER[1], Y. LIU[2], M. WILLIAMS[3], F. IMAI[1], I. PAVLOVA[1], A. LACHHAB[1], R. FRANCOIS[1], S. J. SOBER[3], J. ROGERS[2], Y. YOSHIDA[1];
[1]Burke Neurolog. Inst. | Weill Cornell Med., White plains, NY; [2]Northwestern Univ., Evanston, IL; [3]Emory Univ., Emory Univ., Atlanta, GA

Abstract: Most spinal cord injuries are incomplete, sparing some neural pathways below the injury level. After incomplete cervical spinal cord injury (SCI), axotomized descending axons synapse onto C3-C4 propriospinal neurons (PNs) providing an indirect bypass neuronal pathways to cervical MNs (i.e. bridging the gap). The axons of these PNs, travel in the ventral and lateral white matter and are therefore spared by most incomplete contusive injuries. This intraspinal relay was associated with some spontaneous recovery of voluntary motor control after injury. Yet, anatomical remodeling alone is insufficient to return coordinated motor function. Neuromodulation-based therapies can promote integration of connections between the brain and the spinal cord after injury into functional circuits, and in this way, help to restore a certain degree of motor function. Specifically, optogenetics can provide real-time, selective control of neuronal activity to modulate behavior. In this proposal we aim to leverage opsins with ultra-

high light sensitivity and wireless spinal interface to achieve robust motor recovery in a preclinical model of cervical contusion injury in mice. We surveyed the dynamic reorganization that involves a cardinal class of PNs around and below the lesion site to restore motor output and we tested whether optogenetic stimulation of these neurons can enhance online control of skilled movement. In our preclinical mouse model of SCI, we show that PNs above the injury project past the injury, to connect with motor neurons that drive muscle contraction. Using a novel wireless technology, we further show that we can 'artificially' and 'selectively' modulate the activity of these neurons by cre-dependent expression of ultrapotent opsins. Importantly, we show that our novel optogenetic approach can augment the activity of the spinal pre-motor pathways to restore function by assisting effective control over weakened muscles. Our main outcome measures are changes in key spatial and temporal kinematic features of skilled movement that are largely impaired following SCI (e.g. movement accuracy, velocity profiles and total movement duration) as well as the timing and amplitude of muscle activity (i.e. EMG) of individual muscles and movement synergies in injured mice. While translational applications of optical stimulation have most recently been explored for restoration of vision, seizure control, and treatment of cardiac conditions, our study provides a strong basis for advancing this technology for restoration of motor function following SCI and achieve a better understanding of the neural circuits that mediate functional recovery.

Disclosures: **A. Amer:** None. **Y. Liu:** None. **M. Williams:** None. **F. Imai:** None. **I. Pavlova:** None. **A. Lachhab:** None. **R. Francois:** None. **S.J. Sober:** None. **J. Rogers:** None. **Y. Yoshida:** None.

Poster

PSTR138. Spinal Cord Injury: Biological Repair Strategies

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR138.16/N5

Topic: C.11. Spinal Cord Injury and Plasticity

Support: Science Foundation
Morton Cure Paralysis Foundation
ALARME Foundation
Dr. Miriam and Sheldon G. Adelson Medical Foundation
Wings for Life
Holcim-Stiftung Foundation
Canadian Institutes for Health Research
Human Frontiers in Science Program

Title: Single-cell, multimodal interrogation of spinal cord injury

Authors: *A. Y. TEO[1,2,3], M. A. SKINNIDER[1,2,3,4], M. GAUTIER[1,2,3], C. KATHE[1,2,3], T. H. HUTSON[1,2,3,5], A. LASKARATOS[1,2,3], A. DE COUCY[1,2,3], N. REGAZZI[1,2,3], V. AURELI[1,2,3,6], N. D. JAMES[1,2,3], B. SCHNEIDER[2,7], M. V. SOFRONIEW[8], Q. BARRAUD[1,2,3], J.

BLOCH[1,2,3,6], M. A. ANDERSON[1,2,3,5], J. SQUAIR[1,2,3,6], G. COURTINE[1,2,3,6];
[1]Defitech Ctr. for Interventional Neurotherapies (.NeuroRestore), Swiss Federal Inst. of Technol. (EPFL), of clinical neuroscience, Lausanne Univ. Hosp. (CHUV) and Univ. of Lausanne (UNIL), Lausanne, Switzerland; [2]NeuroX Institute, Sch. of Life Sciences, EPFL, Lausanne, Switzerland; [3]Dept. of Clin. Neuroscience, CHUV and UNIL, Lausanne, Switzerland; [4]Michael Smith Laboratories, Univ. of British Columbia,, Vancouver, BC, Canada; [5]Wyss Ctr. for Bio and Neuroengineering, Geneva, Switzerland; [6]Dept. of Neurosurgery, CHUV and UNIL, Lausanne, Switzerland; [7]Bertarelli Platform for Gene Therapy, EPFL, Lausanne, Switzerland; [8]Dept. of Neurobiology, David Geffen Sch. of Medicine, UCLA, Los Angeles, CA

Abstract: The convoluted nature of spinal cord injury (SCI) lies mainly in the complex cascade of biochemical processes interwoven by distinct responses of various cell types after the initial injury. This progressive cascade is collectively termed the "secondary injury". It involves inflammatory cell infiltration and cytokine release, apoptosis, demyelination, excitotoxicity, ischemia, and the formation of a fibrotic scar with an astrocyte border. Since this is a multi-faceted phenomenon, achieving a complete understanding is no easy feat. Here, we turn to single-cell technology that promises to resolve cell-type-specific molecular programs. We recently generated comprehensive single-cell atlases of the injured mouse spinal cord. The first atlas was built with single-cell transcriptomics (scRNA-seq) data and comprises 400,000 cells spanning 18 experimental conditions. Concurrently, we saw the need to provide a multi-omic context and established a paired scRNA-seq and chromatin accessibility data (scATAC-seq) of 40,000 cells, and spatial transcriptomes of 60,000 spatial barcodes. These were the most comprehensive atlases to date, and have provided us with an unprecedented opportunity to characterize the molecular and cellular makeup of SCI. These atlases have unveiled numerous novel biological principles that underlie the molecular nature of SCI. We leveraged on our multimodal atlas to explore these narratives from a dual perspective at both the transcriptional and epigenomic levels. Our analyses revealed the conserved and divergent neuronal responses to injury; the priming of specific neuronal subpopulations to become circuit-reorganizing neurons after injury; an inherent trade-off between neuronal stress responses and the activation of circuit reorganization programs; and a catastrophic failure to form a tripartite neuroprotective barrier in old mice. Overall, these new findings seek to unravel the underlying molecular and cellular tapestry in SCI and enable a better mechanistic understanding of SCI and consequent effective therapeutics

Disclosures: **A.Y. Teo:** None. **M.A. Skinnider:** None. **M. Gautier:** None. **C. Kathe:** None. **T.H. Hutson:** None. **A. Laskaratos:** None. **A. de Coucy:** None. **N. Regazzi:** None. **V. Aureli:** None. **N.D. James:** None. **B. Schneider:** None. **M.V. Sofroniew:** None. **Q. Barraud:** None. **J. Bloch:** None. **M.A. Anderson:** None. **J. Squair:** None. **G. Courtine:** None.

Poster

PSTR138. Spinal Cord Injury: Biological Repair Strategies

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR138.17/N6

Topic: C.11. Spinal Cord Injury and Plasticity

Support: SNSF 310030_192558 An open-access single-cell atlas of spinal cord injury in zebrafish, rodents and primates

Title: Single-cell interrogation of spinal cord repair across species

Authors: *N. REGAZZI[1,2,3,4], C. KATHE[2,3,4], T. H. HUTSON[5], M. GAUTIER[2,3,4], A. Y. TEO[2,3,4], K. GALAN[2,3,4], S. BORGOGNON[2,3,4], C.-F. V. LATCHOUMANE[2,3,4], S. CETO[2,3,4], S. BRAZ[6], J. NOGUEIRA-RODRIGUES[6], J. M. GIDDAY[7], M. LAWRENCE[7], M. M. SOUSA[6], Q. BARRAUD[2,3,4], M. A. ANDERSON[2,3,4,5], M. SKINNIDER[2,3,4], J. BLOCH[2,3,4,8], G. COURTINE[2,3,4,8], J. SQUAIR[2,3,4,8];
[1]Brain Mind Institute, École Polytechnique Fédérale De Lausan, Geneva, Switzerland; [2]Defitech Ctr. for Interventional Neurotherapies (.NeuroRestore), Swiss Federal Inst. of Technol. (EPFL), of clinical neuroscience, Lausanne Univ. Hosp. (CHUV) and Univ. of Lausanne (UNIL), Lausanne, Switzerland; [3]NeuroX Institute, Sch. of Life Sciences, EPFL, Lausanne, Switzerland; [4]Dept. of Clin. Neuroscience, CHUV and UNIL, Lausanne, Switzerland; [5]Wyss Ctr. for Bio and Neuroengineering, Geneva, Switzerland; [6]i3S, Univ. of Porto, Porto, Portugal; [7]Virscio, Saint Kitts, Saint Kitts and Nevis; [8]Neurosurg. Department, Univ. Hosp. Lausanne (CHUV), Lausanne, Switzerland

Abstract: A SCI triggers a complex and progressive cascade of secondary injuries involving inflammatory cell infiltration and cytokine release, apoptosis, demyelination, excitotoxicity, ischemia, and the formation of a glial scar. How the individual types and subtypes of cells of the central nervous system coordinate to mediate this response remains incompletely understood. Moreover, most of our current knowledge relies on rodent models of SCI, but therapies showing promise in rodents have not translated into successful clinical trials. It is therefore important to better understand the differences between low vertebrate, non-primate mammal and primate models in their molecular responses to injuries and treatments, and to exploit this knowledge to conceive new therapeutic strategies that target mechanisms relevant to the human spinal cord. Here, our goal is to leverage single-nucleus RNA-sequencing (snRNA-seq), to create a freely-available Atlas of the cell type-specific, time-dependent molecular responses to various SCI severities in mouse and rat; in zebrafish, spiny mice, and neonatal mice, which all show varying degrees of spontaneous regeneration; and in primate models, wherein regeneration fails. We complemented this atlas with high-precision kinematic recordings that demonstrated the expected spontaneous behavioural recovery or permanent paralysis in each species, and histological validations with immunohistochemistry and 3D imaging that confirmed the presence or absence of spontaneous axonal regeneration. This cross-species, single-cell molecular atlas of SCI will provide an unprecedented, publicly available resource to improve our understanding of the molecular pathogenesis of SCI. Comparative analyses will reveal biological distinctions between fish, rodent and primate responses to SCI that may have precluded translation of promising preclinical therapies thus far, while simultaneously opening new avenues for targeting molecular mechanisms involved in regeneration.

Disclosures: N. Regazzi: None. **C. Kathe:** None. **T.H. Hutson:** None. **M. Gautier:** None. **A.Y. Teo:** None. **K. Galan:** None. **S. Borgognon:** None. **C.V. Latchoumane:** None. **S. Ceto:** None. **S. Braz:** None. **J. Nogueira-Rodrigues:** None. **J.M. Gidday:** None. **M.**

Lawrence: None. **M.M. Sousa:** None. **Q. Barraud:** None. **M.A. Anderson:** None. **M. Skinnider:** None. **J. Bloch:** None. **G. Courtine:** None. **J. Squair:** None.

Poster

PSTR138. Spinal Cord Injury: Biological Repair Strategies

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR138.18/N7

Topic: C.11. Spinal Cord Injury and Plasticity

Support: Defitech Foundation
Wings for Life
Riders4Riders
Wyss Center for Bio and Neuroengineering
Swiss National Science Foundation PZ00P3_185728 and PZ00P3_208988
Morton Cure Paralysis Foundation
the ALARME Foundation
the Dr. Miriam and Sheldon G. Adelson Medical Foundation
Holcim-Stiftung Foundation
Canadian Institutes for Health Research

Title: Defining the requirements for biological repair and restoration of neurological function across the spectrum of spinal cord injury severities

Authors: *A. DE COUCY[1,2,3,4], J. SQUAIR[2,3,4], M. MILANO[2,3,4], M. GAUTIER[2,3,4], M. SKINNIDER[2,3,4], N. D. JAMES[2,3,4], N. CHO[2,3,4], A. LASNE[2,3,4], C. KATHE[2,3,4], T. H. HUTSON[2,3,4,5], S. L. CETO[2,3,4], L. BAUD[2,3,4], K. GALAN[2,3,4], V. AURELI[2,3,4], A. LASKARATOS[2,3,4], Q. BARRAUD[2,3,4], T. J. DEMING[6], J. BLOCH[2,3,4], R. E. KOHMAN[5], B. L. SCHNEIDER[7], Z. HE[8], M. V. SOFRONIEW[9], G. COURTINE[2,3,4], M. A. ANDERSON[2,3,4,5];
[1]Brain Mind Institute, École Polytechnique Fédérale De Lausan, Geneva, Switzerland; [2]Defitech Ctr. for Interventional Neurotherapies (.NeuroRestore), Swiss Federal Inst. of Technol. (EPFL), of clinical neuroscience, Lausanne Univ. Hosp. (CHUV) and Univ. of Lausanne (UNIL), Lausanne, Switzerland, Geneva, Switzerland; [3]NeuroX Institute, Sch. of Life Sciences, EPFL, Lausanne, Switzerland; [4]Dept. of Clin. Neuroscience, CHUV and UNIL, Lausanne, Switzerland; [5]Wyss Ctr. for Bio and Neuroengineering, Geneva, Switzerland; [6]Departments of Bioengineering, Chem. and Biochemistry, Univ. of California, Los Angeles, CA; [7]Bertarelli Platform for Gene Therapy, EPFL, Geneva, Switzerland; [8]Children's Hosp Boston, Boston, MA; [9]UCLA Schl Med., Los Angeles, CA

Abstract: The clinical challenge posed by spinal cord injury (SCI) remains a significant and unresolved issue. Engineering a treatment for SCI will require biological repair interventions that restore connectivity through or around lesions. Using a combination of advanced neuroanatomical viral tract tracing, single-nucleus RNA sequencing, bioinformatic analyses, behavioral analyses, and in situ validation, we identified a mechanism-based, cell-type specific

gene therapy strategy that is able to induce the regeneration of molecularly-defined subpopulations of spinal cord neurons and guides them to reconnect with their natural target region. This regenerative strategy reverses paralysis following an anatomically complete SCI. However, the majority of human SCI are anatomically incomplete and it remains to be determined how regenerating axons will interact with the distinct lesion compartments of different severities of SCI lesions. Therefore, we aim to dissect the requirements to achieve biological repair and restoration of function across a compendium of SCI lesion models of varying severities. Together, these findings will lay the groundwork for the creation of gene therapy techniques that are suitable for clinical use to repair the injured human spinal cord across a variety of injury severities.

Disclosures: **A. de Coucy:** None. **J. Squair:** None. **M. Milano:** None. **M. Gautier:** None. **M. Skinnider:** None. **N.D. James:** None. **N. Cho:** None. **A. Lasne:** None. **C. Kathe:** None. **T.H. Hutson:** None. **S.L. Ceto:** None. **L. Baud:** None. **K. Galan:** None. **V. Aureli:** None. **A. Laskaratos:** None. **Q. Barraud:** None. **T.J. Deming:** None. **J. Bloch:** None. **R.E. Kohman:** None. **B.L. Schneider:** None. **Z. He:** None. **M.V. Sofroniew:** None. **G. Courtine:** None. **M.A. Anderson:** None.

Poster

PSTR138. Spinal Cord Injury: Biological Repair Strategies

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR138.19/N8

Topic: C.11. Spinal Cord Injury and Plasticity

Support: NIH/NS115759
Paul Kalmanovitz Central Nervous System Repair Research Program
Department of Veterans Affairs Polytrauma and Traumatic Brain Injury Rehabilitation Research Fellowship

Title: A clinically relevant anti-Nogo-A antibody results in functional recovery after spinal cord injury in adult rats

Authors: *S.-Y. TSAI[1], B. E. POWERS[1], J. A. SCHREIBER[1], J. R. LAGHI[1], S. TON[1], E. A. SCHAEFFER[2], D. G. WALLACE[2], R. P. NOCKELS[3], M. E. SCHWAB[5], G. L. KARTJE[1,4]; [1]Res. Service, Edward Hines Jr VA Hosp., Hines, IL; [2]Dept. of Psychology, Northern Illinois Univ., Dekalb, IL; [3]Dept. of Neurolog. Surgery, [4]Dept. of Mol. Pharmacol. and Neurosci., Loyola Univ. Chicago Hlth. Sci. Div., Maywood, IL; [5]NovaGo Therapeut. Inc, Schlieren, Switzerland

Abstract: Spinal cord injuries (SCI) often result in severe functional deficits with currently no available treatment to aid in functional recovery other than rehabilitative therapies that have real but limited success. Numerous studies from our and others' laboratories have reported that treatment with a murine antibody directed against the neurite growth inhibitory protein Nogo-A

resulted in improved functional outcome and regeneration of corticospinal tract (CST) axons after SCI in adult rats, mice and monkeys. To translate these results to human patients, we tested the efficacy of recently developed, fully human antibodies against Nogo-A in a rat model of SCI. Under isoflurane anesthesia, Long Evans adult male rats underwent injury at spinal level T-10 using fine iridectomy scissors to produce a dorsal/dorsolateral hemisection and interrupt the major and minor corticospinal tracts. Using Alzet osmotic minipumps, a catheter was threaded subdurally through an opening at L2 and placed just caudal to the lesion for two weeks of antibody delivery, either human anti-Nogo-A antibody or control antibody. Functional recovery was assessed weekly using the BBB open field locomotor score and the narrow beam task. At eight weeks post-SCI, the hindlimb motor cortex was injected with the anterograde tracer BDA to determine regeneration in the CST. Our behavioral results show that rats treated with the human anti-Nogo-A antibody had significantly improved outcomes in both the BBB score and the narrow beam task when compared to rats treated with control antibody. Anatomical analysis of the spinal cord to determine regeneration or plasticity of spinal pathways which may contribute to this behavioral recovery is currently underway. In conclusion, our results support the efficacy of intrathecally applied antibodies against Nogo-A, including human antibodies, for the treatment of SCI. Such antibodies can then be moved forward into clinical trials.

Disclosures: S. Tsai: None. **B.E. Powers:** None. **J.A. Schreiber:** None. **J.R. Laghi:** None. **S. Ton:** None. **E.A. Schaeffer:** None. **D.G. Wallace:** None. **R.P. Nockels:** None. **M.E. Schwab:** E. Ownership Interest (stock, stock options, royalty, receipt of intellectual property rights/patent holder, excluding diversified mutual funds); NovaGo Therapeutics Inc. **G.L. Kartje:** None.

Poster

PSTR138. Spinal Cord Injury: Biological Repair Strategies

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR138.20/O1

Topic: C.11. Spinal Cord Injury and Plasticity

Support: CSCR20IRG003
CSCR23ERG002

Title: Development of guanine deaminase inhibitors for the treatment of neuropathic pain

Authors: *K. R. LANGE, Jr[1,2], A. M. TSYMBAL[3], N. K. SINGH[2], J. Y. ROBERGE[3], S. ELKABES[4], B. L. FIRESTEIN[2];
[2]Cell Biol. and Neurosci., [3]Mol. Design and Synthesis Core, [1]Rutgers Univ., Piscataway, NJ; [4]Dept. of Neurosurg., New Jersey Med. Sch., Newark, NJ

Abstract: The secondary phase of spinal cord injury (SCI) occurs when disturbances to molecular signaling pathways contribute to accelerated tissue damage after initial trauma. The majority of SCI patients experience neuropathic pain after injury, and current treatments are ineffective, or in the case of opioids, have unwanted addictive properties. Work from our

laboratory demonstrates that inhibition of the guanine deaminase with a small molecule compound, B9, decreases mechanical pain sensitivity in a mouse model of SCI. Additionally, treatment of spinal cord neurons in culture with B9 preserves neuronal viability after glutamate-induced toxicity. However, due to the low potency of B9, there is a need to expand the library of guanine deaminase inhibitors to those with better potency. Using both computational assays, such as protein-ligand molecular docking experiments, and biochemical assays, such as protein thermal shift and xanthine detection, we rapidly screened a library of B9-like compounds to identify compounds with higher potency than B9. Our experiments were successful, and we have identified several molecules that both directly bind to and inhibit guanine deaminase in a manner similar to or better than the B9. The eventual impact of this work is to develop future generations of guanine deaminase inhibitors that may serve as treatments for neuropathic pain.

Disclosures: K.R. Lange: None. **A.M. Tsymbal:** None. **N.K. Singh:** None. **J.Y. Roberge:** None. **S. Elkabes:** None. **B.L. Firestein:** None.

Poster

PSTR138. Spinal Cord Injury: Biological Repair Strategies

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR138.21/O2

Topic: C.11. Spinal Cord Injury and Plasticity

Support: NIH Grant R01

Title: Quantification of locomotor recovery following spinal cord contusion in rats after macrophage depletion

Authors: *G. X. BROWN;
Univ. of Kentucky, Lexington, KY

Abstract: Quantification of locomotor recovery following spinal cord contusion in rats after macrophage depletion

Authors
G. X. Brown[1], R. Kumari[1], A. N. Stewart[1], F. S. Franca[1], S. Kaur[1], W. M. Bailey[1], J. C. Gensel[1];
[1]Spinal Cord and Brain Injury Research Center and Dept. Physiol., Univ. of Kentucky, College of Medicine, Lexington, KY, United States

Disclosures
G. X. Brown: None. **R. Kumari:** None. **A. N. Stewart:** None. **F. S. Franca:** None. **S. Kaur:** None. **W. M. Bailey:** None. **J. C. Gensel:** None.

Abstract
Spinal cord injury (SCI) induces secondary cascades of damage that go beyond the original site of injury. Macrophage-mediated inflammation is one of the accepted mechanisms of secondary degeneration seen after SCI. Specifically, monocyte-derived macrophages infiltrate into the injury site acutely after injury and are associated with damaging surrounding neurons and glial

cells. To further understand the role these leukocytes play in secondary degeneration in SCI, we selectively depleted peripheral macrophages at times of peak inflammation in a rat model of SCI. We utilized 10–12-week-old female Wistar rats that received T9 spinal cord contusion injury (175 kdyn) to model clinical SCI. To deplete the peripheral macrophages, rats received intravenous injections of vehicle or liposome-encapsulated clodronate (2 mL of 7 mg/mL anionic) starting 24-hr after injury, then at 3- and 6-days post-injury (dpi). This method was previously validated and significantly reduces intraspinal macrophage infiltration at 7dpi. To assess behavioral recovery following SCI the horizontal ladder walking test was employed (at baseline, 4 weeks, and 8 weeks post-injury). Data is midway through analysis to comment on behavioral recovery following SCI. Observations from this study will be discussed at the conference. We aim to show how macrophage depletion procedure has a selective nature with efficacy when administered after a SCI, and that cell-specific immunomodulation treatment could be a useful therapy.

Disclosures: G.X. Brown: None.

Poster

PSTR139. Pain Models and Mechanisms

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR139.01/O3

Topic: D.02. Somatosensation – Pain

Support: NIH Grant R01 NS0655926 (TJP)
Robert A. Welch Foundation (F-0652)
NIH Grant 1 S10 OD021508-01 (SFM)

Title: Targeting the sigma-2 receptor/TMEM97 for treating neuropathic pain

Authors: *M. YOUSUF[1], J. J. SAHN[4], H. YANG[5], E. DAVID[1], S. SHIERS[6], D. ROYER[1], C. GARCIA[2], J. ZHANG[1], V. HONG[3], A. AHMAD[7], B. J. KOLBER[8], D. LIEBL[9], S. MARTIN[4], T. J. PRICE[10];
[2]Neurosci., [1]Univ. of Texas at Dallas, Richardson, TX; [3]Univ. of Texas at Dallas, Dallas, TX; [4]Chem., [5]Univ. of Texas at Austin, Austin, TX; [7]Brain and Behavioral Sci., [6]Univ. of Texas At Dallas, Richardson, TX; [8]Neurosci., Univ. of Texas at Dallas, Dept. of Neurosci., Richardson, TX; [9]Univ. Miami, Miami, FL; [10]Sch. of Behavioral and Brain Sci., Univ. of Texas At Dallas Neurosci. Undergraduate Program, Richardson, TX

Abstract: Neuropathic pain is a major medical problem that is poorly treated with existing therapeutics. The Sigma 2 receptor (σ_2R) was described pharmacologically more than three decades ago, but its molecular identity remained obscure until recently when it was identified as transmembrane protein 97 (TMEM97). We and others have shown that σ_2R/TMEM97 ligands produce analgesia in mouse neuropathic pain models with a time course wherein analgesic onset is 24 hours following dosing. We sought to understand this unique anti-neuropathic pain effect

by addressing two key questions: do these σ_2R/TMEM97 compounds act selectively via the receptor, and what is their downstream mechanism on nociceptive neurons. Using male and female conventional knockout (KO) mice for *Tmem97*, we find that a novel σ_2R/TMEM97 binding compound, FEM-1689, requires the presence of the gene to produce analgesia in the spared nerve injury model in mice. Using primary mouse dorsal root ganglion (DRG) neurons, we demonstrate that FEM-1689 inhibits the integrated stress response and promotes neurite outgrowth via a σ_2R/TMEM97-specific action. We extend the clinical translational value of these findings by showing that FEM-1689 reduces ISR and p-eIF2α levels in human sensory neurons and that it alleviates the pathogenic engagement of ISR by methylglyoxal. We also demonstrate that σ_2R/TMEM97 is expressed in human nociceptors and satellite glial cells. These results validate σ_2R/TMEM97 as a promising target for further development for the treatment of neuropathic pain. Our findings demonstrate that σ_2R/TMEM97 targeting with modulators creates analgesia in a mouse model via a specific action on the receptor. We also identify a potential mechanism of action, ISR inhibition, that links the receptor to cellular signaling events that have preclinical and clinical validation for pain relief. Our work suggests that σ_2R/TMEM97 can be selectively engaged by specific small molecules to produce ISR inhibition in a subset of cells that are critical for neuropathic pain. σ_2R/TMEM97-targeted therapeutics thus have the potential to offer effective pain relief without the side effects associated with currently available neuropathic pain medicines.

Disclosures: **M. Yousuf:** E. Ownership Interest (stock, stock options, royalty, receipt of intellectual property rights/patent holder, excluding diversified mutual funds); Founder of NuvoNuro. **J.J. Sahn:** E. Ownership Interest (stock, stock options, royalty, receipt of intellectual property rights/patent holder, excluding diversified mutual funds); Founder of NuvoNuro. **H. Yang:** None. **E. David:** None. **S. Shiers:** None. **D. Royer:** None. **C. Garcia:** None. **J. Zhang:** None. **V. Hong:** None. **A. Ahmad:** None. **B.J. Kolber:** None. **D. Liebl:** None. **S. Martin:** E. Ownership Interest (stock, stock options, royalty, receipt of intellectual property rights/patent holder, excluding diversified mutual funds); Founder of NuvoNuro. **T.J. Price:** E. Ownership Interest (stock, stock options, royalty, receipt of intellectual property rights/patent holder, excluding diversified mutual funds); Founder of NuvoNuro.

Poster

PSTR139. Pain Models and Mechanisms

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR139.02/O4

Topic: D.02. Somatosensation – Pain

Support: AEI/10.13039/501100011033/PID2019-108194RB-100
PROMETEO/2021/031
GRISOLIA/2015/034
PID2019-108194RB-I00
Severo Ochoa Programme for Centres of Excellence in R&D ref. SEV-2017-0723

Title: Role of TRPA1 ion channel and its modulation by Sigma-1 receptor in the prevention of chemotherapy-induced peripheral neuropathy

Authors: A. MARCOTTI[1], J. FERNÁNDEZ-TRILLO[2], A. GONZALEZ[3], M. VIZCAÍNO[2], P. ROS-ARLANZÓN[2], L. ROMERO[4], J. VELA[4], A. GOMIS[2], F. VIANA[2], ***E. DE LA PEÑA GARCIA**[2];
[1]Inst. de Farmacología Exptl. de Córdoba (IFEC), CONICET, Córdoba, Argentina; [2]Inst. De Neurociencias UMH-CSIC, San Juan De Alicante. Alicante, Spain; [3]Div. of Mol. Neurobiology, Dept. of Med. Biochem. and Biophysics, Karolinska Institutet, Stockholm, Sweden; [4]Parc Científic de Barcelona, WeLab Barcelona, Barcelona, Spain

Abstract: Chemotherapy-induced peripheral neuropathy (CIPN) is a frequent side effect of the treatment with different neurotoxic chemotherapeutics, such as platinum derivatives (cisplatin, oxaliplatin, carboplatin) and taxanes (paclitaxel, docetaxel), which are essential in the treatment of different tumors. In the case of patients receiving oxaliplatin, a key drug in the management of colorectal cancer, neuropathy presents as paresthesias, sensory ataxia due to loss of proprioception, mechanical and thermal allodynia, and pain in both hands and feet as in the oral cavity. In many patients, the symptoms are very disabling, making it necessary to reduce the dose of chemotherapeutics, compromising its effectiveness and patient survival. On many occasions, these symptoms not only present acutely while the treatment lasts, but also persist, resulting in chronic pain. TRPA1 is a polymodal, non-selective cation channel expressed in nociceptors, activated by physical stimuli and cellular stress products. TRPA1 has been linked to different neuropathic conditions, including CIPN. Sigma-1 receptor is a ligand-regulated chaperone residing at mitochondria-associated endoplasmic reticulum membranes, and expressed in many tissues, including peripheral sensory neurons. Sigma-1 receptors can translocate to the plasma membrane, regulating the expression and function of many ion channels. In a mice model of oxaliplatin neuropathy (3 i.p. injections, 6 mg/kg), we found that TRPA1 is involved in the development of mechanical and thermal hypersensitivity. Notably, the systemic treatment with S1RA, a selective Sigma-1 receptor antagonist, prevented the development of the painful symptoms in these mice. In calcium imaging studies at cellular level, the incubation of cultured DRG primary sensory neurons with S1RA prevented oxaliplatin-induced TRPA1 sensitization. Using biochemical and biophysical approaches in HEK293-TRPA1 transfected cells we demonstrate that TRPA1 inhibition by S1RA depends of Sigma-1 receptor expression and is not exerted directly on TRPA1 channels. We also demonstrate that S1RA impairs the formation of TRPA1-Sigma-1 receptor complexes, resulting in reduced TRPA1 expression at the plasma membrane. Altogether, these findings provide a mechanistic understanding of the role of Sigma-1 receptor inhibitors in the alleviation of painful CIPN by oxaliplatin and suggest new strategies for its prevention and treatment. All experimental procedures were carried out according to Spanish Royal Decree 53/2013.

Disclosures: A. Marcotti: None. **J. Fernández-Trillo:** None. **A. Gonzalez:** None. **M. Vizcaíno:** None. **P. Ros-Arlanzón:** None. **L. Romero:** None. **J. Vela:** None. **A. Gomis:** None. **F. Viana:** None. **E. De La Peña Garcia:** None.

Poster

PSTR139. Pain Models and Mechanisms

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR139.03/O5

Topic: D.02. Somatosensation – Pain

Support: Ministerio de Ciencia e Innovación
AEI/10.13039/501100011033/PID2019-108194RB-100 co-financed by the European Regional Development Fund (ERDF),
Generalitat Valenciana PROMETEO/2021/031
Severo Ochoa Program for Centers of Excellence in R&D SEV-2017-0723

Title: Piezo2 mediates mechanical sensitivity in MRGPRD nociceptors

Authors: J. FERNANDEZ-TRILLO, F. VIANA, ***A. GOMIS**;
Inst. de Neurociencias, Univ. Miguel Hernandez-CSIC, San Joan D'Alacant. Alicante, Spain

Abstract: Expression of MRGPRD defines a population of non-peptidergic nociceptors that mediate mechanical pain in physiological and pathological conditions. Piezo2 is an ion channel that transduces low-threshold tactile and proprioceptive mechanical stimuli. The participation of Piezo2 in mechanical allodynia has also been suggested. Transcriptomic studies show that Piezo2 is expressed in MRGPRD neurons, however, its role in this population has not been completely elucidated. Thus, we tried to address what is the function of Piezo2 in MRGPRD nociceptive neurons and its possible role in nerve injury-induced mechanical hyperalgesia. First we characterized MRGPRD neurons in mouse dorsal root ganglia (DRG) cultures. Using calcium imaging, we distinguished two subpopulations; one responding to β-alanine and thought to correspond to polymodal nociceptors, and another insensitive to β-alanine previously described as pure mechanonociceptors. Whole-cell patch-clamp experiments revealed that about 50 % neurons from both subpopulations displayed mechanically activated currents with different inactivation kinetics. When similar recordings were performed in cells obtained from a conditional KO mouse lacking Piezo2 in MRGPRD neurons, almost all mechanically activated currents disappeared, suggesting that they are mediated by Piezo2. Behavioral experiments in a model of neuropathic pain (sciatic nerve chronic constriction injury; CCI) showed that the mechanical hypersensitivity that is observed in Piezo2 WT mice, is reduced when Piezo2 is removed from MRGPRD neurons, especially in response to high intensity mechanical stimuli that are more likely activating nociceptors.Finally, using *in vivo* calcium imaging, an increase in the activity of GCamp6f expressing MRGPRD neurons in the L4 DRG (mainly innervated by the sciatic nerve) in response to mechanical noxious stimuli applied to the hind paw was observed after CCI. Strikingly, this increase in activity was abolished in Piezo2 cKO mice. Moreover, the number of MRGPRD neurons responding to mechanical stimulation was reduced in the absence of Piezo2. Altogether, these results strongly suggest that Piezo2 is important for the mechanical sensitivity of MRGRP neurons, both in physiological and neuropathic pain conditions.

Disclosures: J. Fernandez-Trillo: None. **F. Viana:** None. **A. Gomis:** None.

Poster

PSTR139. Pain Models and Mechanisms

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR139.04/O6

Topic: D.02. Somatosensation – Pain

Support: R01NS111553
RFNS113881

Title: Effect of intrathecal NIS-lncRNA antisense oligonucleotides on neuropathic pain caused by nerve trauma, chemotherapy, or diabetes

Authors: *D. SHARMA[1], C.-H. WEN[1], T. BERKMAN[1], S. DU[1], Y. TAO[2];
[1]New Jersey Med. Sch. Rutgers Univ., Newark, NJ; [2]Rutgers Univ., Rutgers New Jersey Med. Sch., Newark, NJ

Abstract: Neuropathic pain, a complex and chronic neurological disorder, affects 7-10% of the general population in the United States. Unfortunately, current treatments for this disorder are often ineffective and/or cause severe side effects. Recent research has shown that abnormal changes in nerve injury-associated genes in the dorsal root ganglion (DRG) play a critical role in the development of neuropathic pain. Our findings indicate that blocking the increased expression of nerve injury-specific long non-coding RNA (NIS-lncRNA) in injured DRG through DRG microinjection of NIS-lncRNA siRNA or generation of NIS-lncRNA knockdown mice can mitigate neuropathic pain. However, these strategies are impractical in clinics. The antisense oligonucleotides (ASOs) are the FDA-approved strategy in the management of neurological diseases. In this study, we reported that intrathecal injection of NIS-lncRNA ASOs on day 7 after chronic constriction injury (CCI) of unilateral sciatic nerve, the fourth lumbar spinal nerve ligation (SNL), or intraperitoneal injection of paclitaxel or on day 14 after intraperitoneal injection of streptozotocin attenuated mechanical allodynia, heat hyperalgesia, cold hyperalgesia, and ongoing nociceptive responses without changing basal or acute nociceptive responses. Intrathecal NIS-lncRNA ASOs also blocked the increases in the levels of NIS-lncRNA and C-C chemokine ligand 2 (CCL2) in the ipsilateral L3 and/or L4 DRGs and hyperactivities of neurons and astrocytes in the ipsilateral L3 and/or L4 spinal cord dorsal horn in these neuropathic pain models. Our findings further validate the role of NIS-lncRNA in trauma-, chemotherapy-, or diabetes-induced neuropathic pain and demonstrates the potential clinical application of NIS-lncRNA ASOs for neuropathic pain management.

Disclosures: D. Sharma: None. **C. Wen:** None. **T. Berkman:** None. **S. Du:** None. **Y. Tao:** None.

Poster

PSTR139. Pain Models and Mechanisms

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR139.05/O7

Topic: D.02. Somatosensation – Pain

Support: R01NS111553
RENS113881

Title: A sensory neuron-specific long noncoding RNA reduces neuropathic pain by rescuing KCNN1 expression

Authors: *L. ZHANG, B. WANG, X. FENG, Y. TAO;
New Jersey Med. Sch., Newark, NJ

Abstract: Nerve injury-induced dysregulation of pain-associated gene expression in dorsal horn ganglion (DRG) plays a critical role in molecular mechanisms underlying neuropathic pain. Long noncoding RNAs (lncRNAs) are key regulators of gene transcription and translation. We recently identified a novel and native lncRNA specifically responded to peripheral nerve injury in DRG, named sensory neuron-specific lncRNA (*SS-lncRNA*). Our preliminary data demonstrated that nerve injury-induced downregulation of *SS-lncRNA* in injured DRG neurons contributed to neuropathic pain development and maintenance. However, the mechanism underlying this phenomenon is still elusive. Here, we showed that rescuing *SS-lncRNA* downregulation reversed a decrease of the calcium-activated potassium channel subfamily N member 1 (KCNN1) in injured DRG and alleviated nerve injury-induced nociceptive hypersensitivity. Mimicking nerve injury-induced DRG *SS-lncRNA* downregulation reduced the expression of KCNN1, decreased total potassium currents and afterhyperpolarization currents and increased excitability in DRG neurons. Mechanistically, downregulated *SS-lncRNA* resulted in the reductions between its binding to *Kcnn1* promoter and heterogeneous nuclear ribonucleoprotein M (hnRNPM), consequent recruitment of less hnRNPM to the *Kcnn1* promoter and silence of *Kcnn1* gene transcription in injured DRG. These findings indicate that *SS-lncRNA* may relieve neuropathic pain through hnRNPM-mediated KCNN1 rescue in injured DRG and offer a novel therapeutic strategy specific for this disorder.

Disclosures: L. Zhang: None. **B. Wang:** None. **X. Feng:** None. **Y. Tao:** None.

Poster

PSTR139. Pain Models and Mechanisms

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR139.06/O8

Topic: D.02. Somatosensation – Pain

Support: R01NS111553
RFNS113881

Title: Fus contributes to nerve injury-induced nociceptive hypersensitivity by activating nf-κb pathway in primary sensory neurons

Authors: G. HAN[1], C.-H. WEN[1], S. WU[1], ***Y. TAO**[2];
[1]Anesthesiol., [2]Rutgers New Jersey Med. Sch., Newark, NJ

Abstract: Dysregulation of pain-associated genes in the dorsal root ganglion (DRG) is considered to be a molecular basis of neuropathic pain genesis. Fused in sarcoma (FUS), a DNA/RNA-binding protein, is a critical regulator of gene expression. However, whether it contributes to neuropathic pain is unknown. This study showed that peripheral nerve injury caused by the fourth lumbar (L4) spinal nerve ligation (SNL) or chronic constriction injury of the sciatic nerve produced a marked increase in the expression of FUS protein in injured DRG neurons. Blocking this increase through microinjection of the adeno-associated virus (AAV) 5 expressing *Fus* shRNA into the ipsilateral L4 DRG mitigated the SNL-induced nociceptive hypersensitivities in both male and female mice. This microinjection also alleviated the SNL-induced increases in the levels of phosphorylated extracellular signal-regulated kinase ½ (p-ERK1/2) and glial fibrillary acidic protein (GFAP) in the ipsilateral L4 dorsal horn. Furthermore, mimicking this increase through microinjection of AAV5 expressing full-length *Fus* mRNA into unilateral L3/4 DRGs produced the elevations in the levels of p-ERK1/2 and GFAP in the dorsal horn, enhanced responses to mechanical, heat and cold stimuli, and induced the spontaneous pain on the ipsilateral side of both male and female mice in the absence of SNL. Mechanistically, the increased FUS activated the NF-κB signaling pathway by promoting the translocation of p65 into the nucleus and phosphorylation of p65 in the nucleus from injured DRG neurons. Our results indicate that DRG FUS contributes to neuropathic pain likely through the activation of NF-κB in primary sensory neurons.

Disclosures: G. Han: None. **C. Wen:** None. **S. Wu:** None. **Y. Tao:** None.

Poster

PSTR139. Pain Models and Mechanisms

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR139.07/P1

Topic: D.02. Somatosensation – Pain

Support: R01NS111553
RFNS113881

Title: Identification of a sensory neuron-specific long noncoding RNA and its role in nuropathic pain

Authors: *X. FENG[1], B. WANG[2], A. BEKKER[2], H. HU[2], Y.-X. TAO[2];
[1]Rutgers, The State Univ. of New Jersey, Livingston, NJ; [2]Rutgers, The State Univ. of New Jersey, Newark, NJ

Abstract: Neuropathic pain is a chronic, refractory clinical condition affecting approximately 6.9-10% of the world population. Maladaptive changes of gene expression in primary sensory neurons are critical for neuropathic pain genesis. Long non-coding RNAs (lncRNAs) regulate gene expression. However, their significance in neuropathic pain remains largely elusive. We recently identified a unique and native lncRNA highly and named sensory neuron-specific lncRNA (SS-lncRNA), for its expression exclusively in dorsal root ganglion (DRG) and trigeminal ganglion. We found that SS-lncRNA is 2.118-kb (containing three exons and poly A tail) in mouse DRG, and 2.454-kb in human DRG (containing three exons and poly A tail). SS-lncRNA was predominantly expressed in small DRG neurons. Peripheral nerve injury down-regulated the expression of SS-lncRNA in injured DRG neurons. Rescuing this down-regulation attenuated the development and maintenance of nerve injury-induced nociceptive hypersensitivity. Mimicking this down-regulation led to neuropathic pain-like behaviors in mice with the absence of peripheral nerve injury. Our findings suggest that DRG SS-lncRNA downregulation is required for neuropathic pain induction and maintenance.

Disclosures: **X. Feng:** None. **B. Wang:** None. **A. Bekker:** None. **H. Hu:** None. **Y. Tao:** None.

Poster

PSTR139. Pain Models and Mechanisms

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR139.08/P2

Topic: D.02. Somatosensation – Pain

Support: U19NS130608

Title: Pathological abnormalities in the human dorsal root ganglion associated with painful diabetic neuropathy

Authors: ***S. SHIERS**[1], K. MAZHAR[2], D. TAVARES-FERREIRA[2], T. PRICE[2];
[1]Neurosci., Univ. of Texas At Dallas, Richardson, TX; [2]Neurosci., The Univ. of Texas at Dallas, Richardson, TX

Abstract: Patients with diabetes mellitus can experience spontaneous sharp and burning pain sensation in the absence of tissue injury. This phenotype is caused by diabetic insults such as reactive oxygen species, excessive polyols, hypoxia, and microvascular issues that lead to neurodegeneration of nociceptive sensory neurons and their axons in the dorsal root ganglia (DRGs). To understand the morphological and molecular underpinnings of how these sensory neurons contribute to diabetic pain, we conducted histological assays and spatial RNA sequencing on human DRGs procured from organ donors with painful diabetic neuropathy (DPN). We note extreme anatomical abnormalities in the DPN DRG samples including the presence of peripherin-positive dystrophic axons, nageotte nodules, and a significant loss of nuclei and SOX10-positive satellite glial cells surrounding sensory neurons. We also find that nageotte nodules are innervated by surviving sensory neurons and form micro neuroma-like

structures within the nodule of non-neuronal cells. To understand what cell-types form these structures and how they may interact with the surviving nociceptors, we conducted spatial RNA sequencing on the DPN DRGs and found many genes for satellite glial cells and Schwann cells localized at these nodules which are abundant in ligands that can interact with receptors found on local immune cells and neurons. These ligand-receptor interactions may represent mechanistic drivers of diabetic neuropathy and evidence new therapeutic intervention opportunities for the treatment of diabetic pain.

Disclosures: **S. Shiers:** None. **K. Mazhar:** None. **D. Tavares-Ferreira:** None. **T. Price:** None.

Poster

PSTR139. Pain Models and Mechanisms

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR139.09/P3

Topic: D.02. Somatosensation – Pain

Support: The Natural Science Foundation of China (Nos. 21732008, and 81771205)
CAMS Innovation Fund for Medical Sciences (CIFMS, Nos. CIFMS-2022-I2M0JB-009, China)
The National Postdoctoral Program for Innovative Talents (No. BX20180053, China)

Title: Rhodojaponin VI indirectly targets Cav2.2 channels via N-ethylmaleimide-sensitive fusion protein to alleviate neuropathic pain

Authors: *T. WANG, F. SUN, Y. FANG, C. MA;
Inst. of Basic Med. Sciences, CAMS&PUMC, Beijing, China

Abstract: Neuropathic pain is a common chronic disease that seriously affects patients' lives and work, but there is currently no particularly effective method for treatment. Therefore, there is an urgent need for drugs with minimal side effects and new therapeutic targets that can alleviate neuropathic pain in clinical practice. Rhododendron VI, a kind of grayanotoxin in *Rhododendron molle*, showed significant analgesic effect in Neuropathic pain model, but its biological target and mechanism of analgesia were not very clear. Because of the reversible effect of rhodojaponin VI and its narrow range of structural modifications, we analyzed the dorsal root ganglion of rats with thermal proteome to determine the protein target of rhodojaponin VI. Using biological and biophysical experiments, we confirmed that the N-Ethylmaleimide-sensitive fusion (NSF) is the key target of rhodojaponin VI. The validation of functional experiments for the first time indicates that NSF promotes the transport of Cav2.2 channels, thereby inducing an increase in Ca^{2+} current intensity, while rhodojaponin VI reverses the effect of NSF. In summary, rhodojaponin VI represents a unique natural analgesic product that targets the Cav2.2 channel through NSF.

Disclosures: **T. Wang:** None. **F. Sun:** None. **Y. Fang:** None. **C. Ma:** None.

Poster

PSTR139. Pain Models and Mechanisms

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR139.10/P4

Topic: D.02. Somatosensation – Pain

Support: NINDS UG3 NS123964 (RCL)
NINDS R21 NS105880 (RCL)
NINDS 3R21NS105880-01S1 (RCL)
DoD W81XWH-19-1-0525 (RCL)

Title: Replication-deficient Herpes Simplex Carbonic anhydrase 8 non-opioid analgesic gene therapy treats chronic osteoarthritis pain by activating sensory neuron kv7 voltage-gated potassium channels

Authors: *G. Z. ZHUANG[1], M. B. KANDEL[3], M. MARZULLI[4], W. F. GOINS[5], J. C. GLORIOSO[6], Y. KANG[3], K. D. SARANTOPOULOS[2], R. C. LEVITT[1];
[2]Dept. of Anesthesiol., [1]Univ. of Miami Miller Sch. of Med., Miami, FL; [3]Anesthesiol., Univ. of Miami MIller Sch. of Med., Miami, FL; [4]Univ. of Pittsburgh Sch. of Med., Pittsburgh, PA; [5]Dept. of Microbiology and Mol. Genet., The Univ. of Pittsburgh Sch. of Med., Pittsburgh, PA; [6]Univ. of Pittsburgh, Pittsburgh, PA

Abstract: Chronic lower extremity osteoarthritis (OA) is a common cause of chronic pain and disability worldwide. Chronic OA pain is inadequately treated making the development of safer analgesics a high priority. In this study we tested the hypothesis that novel JDNI8 replication-defective herpes simplex-1 viral vectors (rdHSV) expressing a carbonic anhydrase-8 analgesic wild-type peptide (vHCA8WT) to treat monoiodoacetate-induced (MIA) chronic OA knee pain. vHCA8WT as compared to CA8 null point mutant (S100P)(vHCA8MT) indicated robust DRG transduction of sensory neurons with vHCA8 (~90% colocalization with advillin; ~60% colocalization with TrkA) using the intra-articular knee joint (KJ) route of administration. vHCA8WT KJ injections inhibited MIA-induced chronic OA mechanical pain by Day 6, returned to baseline mechanical thresholds by Day 13, and exceeded Baseline (anti-nociception) by Day 20 and persisted out to Day 56 as compared to vHCA8MT, which never exceeded baseline mechanical thresholds. vHCA8WT also improved voluntary wheel running, weight bearing and rotarod function. Using allometric conversion, this vHCA8-induced analgesia / antinociception is estimated to equal >100 mg of oral morphine in an average-sized adult. Kv7 channel specific inhibitor XE-991 reversed vHCA8-induced anti-hyperalgesia and analgesia in a dose- and time-dependent manner confirming selective activation of Kv7 voltage-gated potassium channels are involved in these vHCA8 therapeutic effects. These data demonstrate for the first-time local KJ administration of vHCA8WT produces Kv7 channel activation to generate profound analgesia in this MIA-induced chronic OA pain model.

Disclosures: **G.Z. Zhuang:** F. Consulting Fees (e.g., advisory boards); Adolore Biotherapeutics, Inc. **M.B. Kandel:** None. **M. Marzulli:** None. **W.F. Goins:** F. Consulting Fees (e.g., advisory boards); Adolore Biotherapeutics, Inc. **J.C. Glorioso:** F. Consulting Fees (e.g., advisory boards); Adolore Biotherapeutics, Inc. **Y. Kang:** None. **K.D. Sarantopoulos:** None. **R.C. Levitt:** F. Consulting Fees (e.g., advisory boards); Adolore Biotherapeutics, Inc.

Poster

PSTR139. Pain Models and Mechanisms

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR139.11/P5

Topic: D.02. Somatosensation – Pain

Support: NIH Grant NS113991
NIH Grant NS128543

Title: Nociceptor Scaffold Protein Magi-1 as a Novel Therapeutic Target for Osteoarthritis Pain

Authors: *R. RODRIGUEZ[1], A. BHATTACHARJEE[2];
[1]State Univ. of New York, Buffalo, Buffalo, NY; [2]SUNY-Buffalo, Buffalo, NY

Abstract: Persistent pain is one of the most common symptoms associated with osteoarthritis (OA), a major cause of disability in the elderly population. Current treatments such as NSAIDs have failed to efficiently alleviate OA pain in the long term, while reducing adverse effects remains as another challenge for these analgesic drugs. The lack of efficacy of present OA pain medications has aroused an interest in the identification of new drug targets to overcome these barriers. While not very efficient yet, local approaches continue to be a more realistic solution to reduce pain sensitivity in OA patients without causing major side effects. Due to the heterogeneity of OA pain mechanisms, developing novel effective local pain drugs requires the identification of specific targets at joint nociceptors. Our previous studies identified the PDZ and WW domain-containing protein Magi-1 as a key scaffold for voltage-gated ion channels in dorsal root ganglion neurons. Our data showed that Magi-1 regulated Nav1.8 channel trafficking and localization at the plasma membrane, which altered sensory neuronal excitability. Here we aimed to characterize Magi-1 expression in nociceptors that innervate knee joints and determine its role in OA hypersensitivity. Immunostaining analysis showed that Magi-1 and Nav1.8 are both expressed in rodent joint afferent neurons. These results led us to explore the effects of Magi-1 deficiency on pain behavior in the monoiodacetate (MIA) model of knee OA. To achieve this, Magi-1 shRNA-based *in vivo* sciatic nerve transfection was performed to downregulate Magi-1 expression in mouse joint nociceptors after intraarticular MIA injection. The von Frey and dynamic weight bearing assays were used to assess evoked and non-evoked pain behavior in control and Magi-1 shRNA treated mice. Our results showed that Magi-1-deficient mice experienced a reduction in OA pain sensitivity compared to control shRNA treated mice. Additionally, we intraarticularly injected a lipidated peptidomimetic that targets the NaV1.8-Magi1 interaction. We showed that local administration of this peptide decreased pain sensitivity

for a prolonged period of time compared to scrambled control in OA rodents. Altogether, this study seeks to add to the current knowledge on the diversity of OA pain mechanisms, and to identify novel local analgesic targets to treat joint chronic pain.

Disclosures: R. Rodriguez: None. **A. Bhattacharjee:** None.

Poster

PSTR139. Pain Models and Mechanisms

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR139.12/P7

Topic: D.02. Somatosensation – Pain

Support: NIH Grant NS 113991
NIH Grant NS 128543

Title: Inhibiting endocytosis in nociceptors alters MIA-Induced Osteoarthritis pain behavior

Authors: *A. COOPER[1], O. M. JOSHI[2], J. TABMAN[2], R. RODRIGUEZ[3], A. BHATTACHARJEE[4];
[1]State Univ. of New York, Univ. at Buffalo, Buffalo, NY; [2]Universitu at Buffalo, Buffalo, NY; [3]State Univ. of New York, Buffalo, Buffalo, NY; [4]SUNY-Buffalo, Buffalo, NY

Abstract: Osteoarthritis (OA) is a degenerative joint condition that leads to chronic pain and a consequential need for pain relief. Current pain management for OA pain is fraught with efficacy issues and adverse effects. Our previous studies have indicated that inhibition of the nociceptor endocytotic adaptor protein complex 2 (AP2) inhibits neuronal hyperexcitability and inflammatory pain behavior (PMID: 34608164). Here we show the AP2A2 subunit localized to CGRP-containing large dense core vesicles (LDCVs) in human (n=3) and mouse DRG neurons. Immunohistochemistry was also performed on rodent knee joints to demonstrate AP2 co-localization with CGRP in synovial afferents. Furthermore, we demonstrate that pain behavior in arthritis is reduced by local pharmacological inhibition of the nociceptor AP2 complex using a lipidated AP2 peptide inhibitor. Monoiodoacetate (MIA) was used to induce knee joint OA in male and female Sprague-Dawley rats. Four days later, upon verification of pain, a one-time intra-articular injection to the arthritic knee of either the AP2 inhibitor peptide or a scrambled peptide control was administered. Joint pain was assessed via a dynamic weight-bearing (DWB) assay. We also assessed the effects of the peptides on central sensitization via the von Frey method. Pain behavior was monitored over the course of 28 days post MIA-induced OA. DWB analyses showed the typical decrease in weight bearing in the group whose arthritic knees were injected with scrambled peptide; however, an increase in weight bearing, persisting up to 24 days, was observed in the AP2 peptide injected group. Similarly, the group that received the scrambled peptide showed a decrease in paw withdrawal threshold (PWT) when subjected to the von Frey filaments after post MIA-induced OA; whereas the AP2 inhibitor peptide prevented such a change in evoked pain sensitivity. Micro-computed tomography and histological analyses

were performed on MIA injected knee joints as well as the contralateral healthy knee joints at end of 28 days to evaluate the effects on disease progression. Pathological analyses indicated the typical reduction of subchondral bone content and cartilage formation in scrambled peptide-injected MIA-treated joints, however intra-articular AP2 inhibitor peptide injections resulted in a significant retention of bone content and cartilage. This data suggests that inhibition of joint nociceptor endocytosis can decrease OA pain and modify disease.

Disclosures: A. Cooper: None. **O.M. Joshi:** None. **J. Tabman:** None. **R. Rodriguez:** None. **A. Bhattacharjee:** None.

Poster

PSTR139. Pain Models and Mechanisms

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR139.13/P8

Topic: D.02. Somatosensation – Pain

Support: CIHR PJT-173355

Title: Evaluation of TACAN as a new target for treating osteoarthritis pain.

Authors: *A. GILBERT[1], L. S. MIRAUCOURT[5], L. S. RABILLIER[2], M. GEORGIOPOULOS[3], I. COLMEGNA[4], J. OUELLET[3], R. DALLEL[6], C. PEIRS[7], R. SHARIF NAEINI[2];
[1]McGill Univ., MONTREAL, QC, Canada; [3]Surgery, [4]Rheumatology, [2]McGill Univ., Montreal, QC, Canada; [5]Montreal Neurolog. Institute, McGill Univ., Montreal, QC, Canada; [7]Neuro-Dol, [6]Clermont Auvergne Univ., Clermont-Ferrand, France

Abstract: Joint pain is the most prominent symptom of osteoarthritis (OA). Patients with OA experiencemechanical allodynia which is due in large part to a dysfunction in nociceptors. We previouslydemonstrated that during OA, mechanosensitive ion channels, which convert high-intensitymechanical stimuli into electrical signals, become sensitized and are contribute to mechanicalallodynia, where light stimuli are perceived as painful. However, the molecular identity of thesechannels was unknown, which prevented any progress toward improved pain management in OApatients. We recently identified an ion channel expressed in mouse nociceptors, called TACAN,essential to the sensation of mechanical pain. Here, we examined whether TACAN is necessaryto the development of mechanical allodynia in OA. We used behavioural tests to assess primarymechanical allodynia and pain in a preclinical model of knee OA in both male and female mice.We decreased TACAN expression by injecting adeno-associated viral vectors encoding control orTACAN shRNA in the knee capsule. Deletion of TACAN in nociceptors of OA mice significantlydecrease primary mechanical allodynia. In electrophysiology experiments, we characterized thecontribution of TACAN to the mechanosensitivity of both mouse and human nociceptors fromrecently deceased donors incubated with control media or synovial fluid (SF) obtained from OApatients. Mechanically

evoked responses are potentiated following a 24h incubation period withOA-SF: Mean amplitude, percentage of active patch are significantly increased. Furtherinvestigation will assess whether the inflammatory mediators contained in the SF can lead to thesensitization of mechanical responses in human nociceptors via the TACAN channel and modulatepain associated symptoms during OA.

Disclosures: **A. Gilbert:** None. **L.S. Miraucourt:** None. **L.S. Rabillier:** None. **M. Georgiopoulos:** None. **I. Colmegna:** None. **J. Ouellet:** None. **R. Dallel:** None. **C. Peirs:** None. **R. Sharif Naeini:** None.

Poster

PSTR139. Pain Models and Mechanisms

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR139.14/P9

Topic: D.03. Somatosensation – Touch

Support: NSERC RGPIN-2016-05538
CIHR FRN-162179

Title: Investigation of spinal networks underlying plastic changes in nociceptive processing

Authors: *J. CHEUNG[1], M. ZAIN[1], R. BONIN[1,2];
[1]Univ. of Toronto, Toronto, ON, Canada; [2]Univ. of Toronto Ctr. for the Study of Pain, Toronto, ON, Canada

Abstract: Chronic pain is a major global health concern, affecting 20% of the world's population. Capsaicin is the spicy component of chili peppers and is a direct transient receptor potential vanilloid 1 (TRPV1) agonist. Since TRPV1 is a key receptor in pain signaling, capsaicin is often used in the modelling and study of pain in humans and animals. In humans, the application of capsaicin with a nerve blocker has been shown to prevent the development of mechanical sensitization suggesting capsaicin-induced mechanical sensitization is a central process. Furthermore, TRPV1-expressing afferents directly innervate neurons of the superficial dorsal horn of the spinal cord. Therefore, to investigate the mechanisms underlying capsaicin-induced mechanical sensitization, we sought to develop an *ex vivo* spinal cord model. This model consists of the isolation of the lumbar region of the spinal cord from adult mice with sensory roots intact. Afferent terminals were activated by a bath application of capsaicin or by optogenetic techniques. The optogenetic model consists of a lumbar spinal cord explant from TRPV1-channelrhodopsin (ChR2) mice in which all TRPV1-lineage neurons are also channelrhodopsin-positive. Neuronal activation was identified by c-Fos immunofluorescence staining and confocal microscopy and quantified by cell counting. TRPV1 staining of the spinal cord show its localization to the superficial dorsal horn thereby confirming the region of interest used for cell counting. A dose-response study of 0.3, 1, and 3 µM of capsaicin performed suggest bath application of capsaicin may result in an increase in neuronal activation in the superficial

dorsal horn of the spinal cord. The optogenetic approach consisting of blue light stimulation of the lumbar spinal cord for 3 minutes at 2 Hz suggest a potential increase in neuronal activation in the stimulated group compared to baseline. Furthermore, blue light stimulation of spinal cords isolated from TRPV1-ChR2 mice showed a greater magnitude in neuronal activation compared to wild type. Together, these results suggest the spinal explant model allows for the direct assessment of the plastic changes in nociceptive processing in the spinal cord through the direct manipulation and investigation of sensory network function.

Disclosures: J. Cheung: None. **M. Zain:** None. **R. Bonin:** None.

Poster

PSTR139. Pain Models and Mechanisms

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR139.15/P10

Topic: D.03. Somatosensation – Touch

Support: NSERC RGPIN-2016-05538
CIHR FRN-162179

Title: Regulation of mechanical hypersensitivity by amyloid-beta in the mouse spinal dorsal horn

Authors: *L. A. BENNETT, H. ZHANG, R. P. BONIN;
Pharmaceut. Sci., Univ. of Toronto, Toronto, ON, Canada

Abstract: Synaptic plasticity that allows for memory in the brain has mechanistic and functional parallels to synaptic plasticity that occurs between neurons in the spinal dorsal horn. The small peptide, amyloid-beta, is associated with memory loss in Alzheimer's disease but is present at endogenously low concentrations in brains of healthy individuals. We hypothesize that amyloid-beta contributes to synaptic plasticity and sensory processing in the spinal dorsal horn. Our overall aim is to modulate amyloid-beta in the spinal dorsal horn to improve hypersensitivity in pain models.We used Enzyme Linked Immunosorbent Assay (ELISA) to quantify amyloid-beta in the spinal cord during central sensitization, induced via capsaicin, complete Freund's adjuvant (CFA) and spared nerve injury (SNI). We increased amyloid-beta levels transiently via synthetic amyloid-beta and decreased amyloid-beta levels via gamma-secretase inhibitor DAPT. We used a knockout mouse model of the amyloid precursor protein (APP KO) which does not produce amyloid-beta and an APP mouse model that over produces amyloid-beta (TgCRND8). Sensory sensitivity was tested using Von Frey and Hargreaves. Mechanical sensitivity of CFA-injected mice were not impacted by synthetic amyloid-beta or DAPT injection. Interestingly APP KO mice did not respond with a change in mechanical sensitivity after CFA whereas TgCRND8 male mice did show improved mechanical sensitivity. After SNI, both sexes showed improvement in mechanical sensitivity after DAPT injection and after amyloid-beta injection in

females only. Taken together, our results thus far indicate modulation of amyloid-beta may play a role in the mechanical sensitivity attributed to a model of neuropathic pain.

Disclosures: L.A. Bennett: None. **H. Zhang:** None. **R.P. Bonin:** None.

Poster

PSTR139. Pain Models and Mechanisms

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR139.16/Q1

Topic: D.03. Somatosensation – Touch

Support: Canadian Institutes of Health Research FRN 162179
Natural Sciences and Engineering Research Council of Canada RGPIN-2016-05538

Title: Non-ionotropic NMDA receptor signaling reverses pathological pain in a sensory model of reconsolidation.

Authors: *H. ZHANG[1], L. D. RODRIGUEZ-HERNANDEZ[1], A. J. D'SOUZA[1], D. HE[2], M. ZAIN[1], S. W. FUNG[1], L. A. BENNETT[1], R. P. BONIN[1,3,4];
[1]Leslie Dan Fac. of Pharm., [2]Dept. of Anesthesia, Temerty Fac. of Med., [3]Dept. of Cell and Syst. Biol., Univ. of Toronto, Toronto, ON, Canada; [4]Univ. of Toronto Ctr. for the Study of Pain, Toronto, ON, Canada

Abstract: **Objective and rationale:** Chronic, pathological pain is a highly debilitating condition maintained through central sensitization, which shares many parallels with memory formation. Injury leaves a 'pain memory' that can be dynamically regulated and reversed by reactivating sensory pathways. However, the mechanisms underlying synaptic destabilization of spinal 'pain engram' during reactivation (or Reactive destabilization) remain poorly understood. This project aims to explore the signaling cascades that directly contribute to the synaptic depotentiation enabled by reactive destabilization. **Methods and Results:**Using animal pain models, electrophysiological recordings, and western blotting assays, we explored the role of non-ionotropic NMDA receptor (NI-NMDAR) signaling in reactive destabilization. This ion-flux independent NMDA receptor activity could be triggered by nociceptor activity and was necessary and sufficient to reduce mechanical hyperalgesia with long-lasting analgesic effects and can also attenuate dorsal horn LTP. NI-NMDAR signaling could reverse sensitization in different mouse strains and sexes. Furthermore, C-terminal tail of the NMDA receptor GluN1 subunit is coordinating the destabilization of sensitized pain pathway and required ubiquitin-mediated degradation of post-synaptic proteins. We are currently examining the role of NI-NMDAR activity in resolving ongoing pain, particularly in a neuropathic pain model induced by spared nerve injury in both sexes. Pairing intraplantar capsaicin injection with spinal administration of the protein synthesis inhibitor, anisomycin (47 mM, i.t.), one week after injury resulted in a significant but partial (60%) reversal of mechanical hypersensitivity compared to

the vehicle control group. Notably, intrathecal administration of either anisomycin or capsaicin alone did not have any effect on mechanical hypersensitivity. Additionally, we have utilized an NMDA receptor deficiencies patient-derived mouse line with reduced glycine binding affinity to GluN1 subunit of NMDA receptor. This model will provide insights into the necessity of GluN1 subunit in destabilizing sensitized pain pathways, resembling effects of NI-NMDAR activity induced by drugs.**Conclusions:**Our ongoing research aims to contribute to the understanding of the endogenous role of non-ionotropic NMDAR activity in synaptic plasticity and its therapeutic potential for chronic pain conditions. By elucidating the mechanism underlying reactive destabilization in the sensory model of reconsolidation, we hope to uncover new targets for the development of more effective pain treatments.

Disclosures: H. Zhang: None. **L.D. Rodriguez-Hernandez:** None. **A.J. D'Souza:** None. **D. He:** None. **M. Zain:** None. **S.W. Fung:** None. **L.A. Bennett:** None. **R.P. Bonin:** None.

Poster

PSTR140. Interoception

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR140.01/Q2

Topic: F.06. Autonomic Regulation

Support: HHMI

Title: Brain-wide mapping of gut interoception

Authors: *W. CHEN[1], B. JAMES[1], V. RUTTEN[1], S. BANALA[1], S. NARAYAN[1], M. RUBINOV[2], L. LAVIS[1], J. FITZGERALD[1], M. AHRENS[1];
[1]Janelia, HHMI, Ashburn, VA; [2]Vanderbilt Univ., Nashville, TN

Abstract: An organism's sensory environment is not encompassed entirely by the external environment – sensory systems also monitor the state of internal organs, sending this information to the brain in a process known as interoception. It remains unclear how interoception is processed at whole-brain scale in intact neuronal circuits in awake animals. Here we present a holistic system for studying the gut-brain axis in larval zebrafish. By combining whole-brain light-sheet imaging, optical uncaging of compounds in the gut, and sensory and motor input/output recordings, we gain insights into how internal information is encoded in the brain and integrated with external cues. Optically uncaging compounds like glucose and glutamate in the gut enables precise investigation of gut sensory processing without significant movement artifacts. The concurrent delivery of visual optomotor stimuli enables dissecting how internal signals affect known visual and motor circuits, while recording motor signals and constructing a closed-loop virtual reality environment opens the possibility of studying how internal states affect behavior. Gut stimuli evoke activity across the brain, indicating a widespread impact of gut signals. These neurons not only respond to gut stimuli but also receive inputs from other sources, such as visual and motor information, suggesting their integration with various sensory

and motor pathways. Furthermore, the gut-responsive neurons encode different intestinal chemicals in an intermingled manner, implying their ability to represent diverse chemical signals. Chemical release in different parts of the gut leads to differential activation in specific brain regions, suggesting the spatial specificity of gut-brain communication, while the modulation of visual information processing by gut sensation further demonstrates the influence of gut signals on sensory perception. Taken together, we developed a paradigm for studying the gut-brain axis at the scale of the entire larval zebrafish brain, and we map its core architecture and interactions with other sensory and motor systems.

Disclosures: **W. Chen:** None. **B. James:** None. **V. Rutten:** None. **S. Banala:** None. **S. Narayan:** None. **M. Rubinov:** None. **L. Lavis:** None. **J. Fitzgerald:** None. **M. Ahrens:** None.

Poster

PSTR140. Interoception

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR140.02/Q3

Topic: D.04. The Chemical Senses

Support: Boehringer Ingelheim Fonds PhD Fellowship
DIBS Research Incubator Award
Sloan Research Fellowship

Title: Mapping brain-wide responses to gut-mediated signals in larval zebrafish

Authors: ***M. ARINEL**[1], J. ATKINSON[1], D. ECHEVERRI[1], K. M. MATOS-FERNÁNDEZ[1], E. P. DRAGE[2], M. HAWKYARD[3], E. A. NAUMANN[1];
[1]Dept. of Neurobio., Duke Univ., Durham, NC; [2]Dept. of Genet. and Mol. Biol., Univ. of North Carolina, Chapel Hill, Chapel Hill, NC; [3]Aquaculture Res. Inst., Univ. of Maine, Orono, ME

Abstract: Specialized sensory cells in the gut epithelium termed enteroendocrine cells (EECs) have been shown to form synaptic connections with vagal sensory neurons, sending enteric information directly to the brain through the vagus nerve. Yet, we lack basic conceptual insights into the way gut-brain circuits encode sensory information to impact neural activity across the brain. The translucent zebrafish is an ideal model due to its optical accessibility across the entire gut-brain circuitry at single-cell resolution. Here, we demonstrate a method to study the effects of EEC-mediated signals on vagal and brain-wide activity using chemical and optogenetic approaches in larval zebrafish. To map functional responses, we engineered a computer controlled microgavage system to inject nanoliter volumes of distinct stimuli directly into the intestinal lumen of larval zebrafish while performing calcium imaging via volumetric two-photon microscopy. Using precisely timed injections of EEC-activating nutrients directly into the intestinal bulb, we show differential activation of vagal and hindbrain neurons in fish with different feeding experiences, suggesting specific enteric sensory encoding across these brain

areas. Finally, optogenetic photostimulation of EECs along the gut directly implicates these cells in driving neural activity and establishes topographic representations of the gut across the brain.

Disclosures: **M. Arinel:** None. **J. Atkinson:** None. **D. Echeverri:** None. **K.M. Matos-Fernández:** None. **E.P. Drage:** None. **E.A. Naumann:** None.

Poster

PSTR140. Interoception

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR140.03/Q4

Topic: D.04. The Chemical Senses

Support: NIH DP1AT009497
NIH R01DK103703
NIH T32 HL007901
Food Allergy Science Initiative
Japan Society for the Promotion for Science

Title: Enteroendocrine cell lineages that differentially control feeding and gut motility

Authors: *M. HAYASHI[1], J. KAYE[2], N. R. JOSHI[3], E. DOUGLAS[2], F. REIMANN[5], F. M. GRIBBLE[5], S. LIBERLES[4];
[1]Harvard medical school, Boston, MA; [3]Dept. of Cell Biol., [4]Harvard Med. Sch., [2]Harvard Med. Sch., Boston, MA; [5]Univ. of Cambridge, Univ. of Cambridge, Cambridge, United Kingdom

Abstract: Enteroendocrine cells are specialized sensory cells of the gut-brain axis that are sparsely distributed along the intestinal epithelium. The functions of enteroendocrine cells have classically been inferred by the gut hormones they release. However, individual enteroendocrine cells typically produce multiple, sometimes apparently opposing, gut hormones in combination, and some gut hormones are also produced elsewhere in the body. Here, we developed approaches involving intersectional genetics to enable selective access to enteroendocrine cells in vivo in mice. We targeted FlpO expression to the endogenous *Villin1* locus (in *Vil1-p2a-FlpO* knock-in mice) to restrict reporter expression to intestinal epithelium. Combined use of Cre and Flp alleles effectively targeted major transcriptome-defined enteroendocrine cell lineages that produce serotonin, glucagon-like peptide 1, cholecystokinin, somatostatin, or glucose-dependent insulinotropic polypeptide. Chemogenetic activation of different enteroendocrine cell types variably impacted feeding behavior and gut motility. Defining the physiological roles of different enteroendocrine cell types provides an essential framework for understanding sensory biology of the intestine.

Disclosures: **M. Hayashi:** None. **J. Kaye:** None. **N.R. Joshi:** None. **E. Douglas:** None. **F. Reimann:** None. **F.M. Gribble:** F. Consulting Fees (e.g., advisory boards); consultant for Kallyope, Inc. **S. Liberles:** F. Consulting Fees (e.g., advisory boards); consultant for Kallyope, Inc.

Poster

PSTR140. Interoception

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR140.04/Q5

Topic: D.04. The Chemical Senses

Support: Warren Alpert foundation

Title: Par1 expressing subpopulation of vagal c-fibers mediate hdm induced airway hyperreactivity

Authors: *M. PATIL[1], M. SARGEANT[4], K. LURYE[4], N. PAVELKOVA[5], S. HADLEY[4], S.-H. KIM[2], T. TAYLOR-CLARK[3], A. N. AKOPIAN[6];
[1]MPP, [2]Dept. of Mol. Pharmacol. and Physiol., [3]Mol. Pharmacol. & Physiol., Univ. of South Florida, Tampa, FL; [4]MPP, USF, Tampa, FL; [5]MPP, USF, tampa, FL; [6]UT Hlth. Sci. Ctr., San Antonio, TX

Abstract: Patients suffering from allergic asthma have airway hyperreactivity (AHR) which is the main cause of exacerbations. None of the current therapeutics effectively target AHR. The airway afferent/sensory nerves have been recently shown to play a critical role in AHR, but the mechanisms are not well understood. We found that diphtheria toxin-mediated ablation of C-fibers using Nav1.8^{Cre}, caused a significant reduction in HDM induced AHR in mice. Employing two-photon *ex vivo* imaging of the vagal ganglia of Pirt^{Cre};R26-GCaMP6s mice, HDM extract caused a robust activation of majority of naïve airway-specific C-fibers and this activation was via PAR1 receptors. We created sensory neuronal conditional knockout of PAR1 and PAR2 using the Pirt^{Cre} mice crossed with PAR1 and PAR2 floxed mice to generate experimental Pirt^{Cre}/PAR1$^{fl/fl}$ and Pirt^{Cre}/PAR2$^{fl/fl}$ CKO mice. We found that Pirt^{Cre}/PAR1$^{fl/fl}$ (CKO) mice exposed to HDM over a period of 12 days had significantly reduced HDM-induced AHR. Our preliminary two-photon HDM activation studies with PirtCre;R26-GCaMP6s mice has shown that HDM activates most of the vagal C-fibers, but it also activates non-nociceptive A fibers that lack TRPV1. Thus, we observe 3 major vagal afferent subtypes, nodose P2X2/TRPV1+, jugular Tac1/TRPV1+ and TRPV1-. We have generated TRPV1^{Flp} mice which were crossed with either P2X2^{Cre} or Tac1^{Cre} to obtain mice in which nodose, or jugular subtypes can be selectively labeled by appropriate dual Flp-/Cre-sensitive reporters. Importantly, these strategies allow us, for the first time, to selectively label and ablate vagal nodose or jugular C-fiber subtypes independently. Our preliminary evidence has led us to the **novel hypothesis** **that *PAR1 expressing vagal C-fiber afferent subpopulation mediate HDM-induced airway hyperreactivity*.**

Disclosures: M. Patil: None. **M. Sargeant:** None. **K. Lurye:** None. **N. Pavelkova:** None. **S. Hadley:** None. **S. Kim:** None. **T. Taylor-Clark:** None. **A.N. Akopian:** None.

Poster

PSTR140. Interoception

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR140.05/Q6

Topic: D.02. Somatosensation – Pain

Support: Canada Graduate Scholarship - Master's

Title: Nociceptor neurons control vaccine-induced immunity

Authors: *S. GUPTA[1], F. BORRIELLO[3], J.-C. WANG[5], H. MERRISON[6], A. J. DUTTON[6], D. DOWLING[3], C. J. WOOLF[4], O. LEVY[3], S. L. FOSTER[6], S. TALBOT[2,7];
[1]Ctr. for Neurosci., [2]Dept. of Biomed. and Mol. Sci., Queen's Univ., Kingston, ON, Canada; [3]Dept. of Medicine, Div. of Infectious Dis., [4]Children's Hosp. Boston, Children's Hosp. Boston, Boston, MA; [5]Dept. of Pharmacol. and Physiol., Univ. of Montreal, Montreal, QC, Canada; [6]Dept. of Psychiatry, Massachusetts Gen. Hosp. and Harvard Med. Sch., Boston, MA; [7]Dept. of Physiol. and Pharmacol., Karolinska Institutet, Stockholm, Sweden

Abstract: Nociceptors, the sensory neurons that detect noxious stimuli and trigger pain, interact with immune cells to modulate immune responses. The nociceptor-released neuropeptide substance P promotes B cell polarization, antibody class switching to IgE, and IgE release in models of allergic inflammation. In this study, we investigated whether nociceptors respond to vaccine adjuvants and control IgG production and clonal selection in the context of vaccination. We activated and sensitized sensory neurons from mice with vaccines and adjuvants against influenza virus and pneumococcal and meningococcal bacteria in vitro and evaluated influenza vaccine-specific IgG antibody levels in mice with ablated nociceptors. Our results showed that sensory neurons respond to vaccines and exhibit differential activation by various noxious ligands. In mice with ablated nociceptors, IgG2c titers were reduced, while capsaicin-treated mice showed increased IgG titers. These findings suggest a role for nociceptors in maintaining humoral immunity after vaccination. We will further explore how sensory neuron ablation or overactivation affects B-cell trafficking and antibody production in response to mouse vaccination and pathogen challenges. This research provides insights into the role of nociceptor neurons in humoral immune responses during vaccination and has implications for developing more effective vaccines.

Disclosures: S. Gupta: None. **F. Borriello:** None. **J. Wang:** None. **H. Merrison:** None. **A.J. Dutton:** None. **D. Dowling:** None. **C.J. Woolf:** None. **O. Levy:** None. **S.L. Foster:** None. **S. Talbot:** None.

Poster

PSTR140. Interoception

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR140.06/Q7

Topic: D.04. The Chemical Senses

Title: Uncovering Sensory Nerve Functions in White Adipose Tissue

Authors: *G. B. MISHRA[1], M. BLASZKIEWICZ[2], A. TEDESCHI[3], G. GUNSCH[4], J. WILLOWS[5], M. IADAROLA[6], M. IADAROLA[6];
[1]Neurosurg., The Ohio State Univ. Neurosci. Grad. Program, Columbus, OH; [2]Neurosurg., The Ohio State Univ. Col. of Med., Columbus, OH; [3]The Ohio State Univ., The Ohio State Univ., Columbus, OH; [4]Neurosurg., [5]Ohio State Univ., Columbus, OH; [6]Natl. Inst. of Hlth., Bethesda, MD

Abstract: Uncovering Sensory Nerve Functions in White Adipose Tissue
White adipose tissue (WAT) is the primary energy storage site in the body, and is densely innervated by sensory and sympathetic nerves that enable bi-directional communication with the central nervous system. The sympathetic neurotransmitter norepinephrine is the best-studied nerve product in WAT, and is known to stimulate lipolysis, browning (development of brown adipocytes in WAT), thermogenesis, and other metabolically favorable processes. Conversely, WAT sensory nerves secrete neuropeptides like calcitonin gene related peptide (CGRP), a vasodilatory neuropeptide that also impacts lipolysis, but CGRP functions and mechanisms remain vastly understudied, especially in adipose tissues. Circulating CGRP levels are known to be increased in obese humans, but given the beneficial metabolic effects of CGRP, this may be a case of CGRP dysregulation or resistance. Thus, a more comprehensive investigation into CGRP's roles in adipose tissue and the importance of the sensory nerve supply for WAT function and obesity is warranted. First, we identified CGRP distribution in sensory nerves innervating mouse WAT by whole-mount imaging the inguinal WAT depot from sensory nerve reporter (Nav1.8Cre x tdTomato) marked with CGRP. We then observed significant increase in WAT CGRP levels by ELISA 30 minutes after directly delivering 13-HODE, a naturally occurring lipid-based sensory nerve receptor transient potential vanilloid channel agonist, into WAT. However, CGRP levels returned to baseline by 120 minutes after treatment, suggesting CGRP release may be an acute response to incoming stimuli and a consequence of interoceptive response by sensory nerves. Moreover, *in vitro* treatment of pre-adipocytes with recombinant CGRP for 60 minutes upregulated lipolytic pathways, as measured by phosphorylated hormone sensitive lipase levels (pHSL/HSL) western blot. Hence, sensory nerve derived CGRP may directly promote lipolysis in adipocytes. Moreover, using CGRP ELISA, we have observed CGRP levels in WAT are altered with changing energy balance states, as well as by acute versus chronic changes to altered energy balance states. Specifically, with obesity, in both diet-induced and genetic *ob/ob* mice, we see increased WAT CGRP levels, which fits the reports in humans and may be due to chronically elevated tissue lipids. Collectively, CGRP is a novel neuropeptide in adipose that may be relevant for lipolytic stimulation, changes in energy balance statues, and likely contributes to metabolic health.

Disclosures: G.B. Mishra: None. **M. Blaszkiewicz:** None. **A. Tedeschi:** None. **G. Gunsch:** None. **J. Willows:** None. **M. Iadarola:** None. **M. Iadarola:** None.

Poster

PSTR140. Interoception

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR140.07/Q8

Topic: D.02. Somatosensation – Pain

Support: NIH R25
Massachusetts General Hospital Department of Medicine
Howard Hughes Medical Institute and Harvard Medical School

Title: Genetically and morphologically distinct DRG sensory neurons innervate the distal colon and mediate distinct behavioral responses to colonic stimulation

Authors: *A. ABDELAZIZ[1], R. WOLFSON[1,2], S. KUSHNER[1], D. GINTY[1];
[1]Dept. of neurobiology, Harvard Univ., Boston, MA; [2]Massachusetts Gen. Hosp., Boston, MA

Abstract: Abdominal pain is prevalent across many patient populations, yet the underlying pathophysiology is poorly understood. Here we focus on sensory afferents that innervate the distal colon, whose cell bodies reside in dorsal root ganglia (DRG). We hypothesized that subsets of $CGRP^+$ DRG sensory neurons play a nociceptive role in the distal colon. We used newly available mouse somatosensory neuron genetic tools, anatomy, and behavior to assess the properties and functions of DRG sensory neurons innervating the distal colon. A sparse labeling strategy was used to assess the morphology and colon innervation patterns of several DRG subtypes. Both $Bmpr1b^+$ Aδ high threshold mechanoreceptors (HTMRs), which are a subset of $CGRP^+$ neurons, and $TrkB^+$ Aδ low threshold mechanoreceptors (LTMRs) innervate the myenteric plexus layer of the distal colon, however they are morphologically different. $Bmpr1b^+$ Aδ-HTMRs form simple endings whereas $TrkB^+$ Aδ-LTMRs form elaborate endings. The $Sstr2^+$ C-HTMRs, another subset of $CGRP^+$ neurons, terminated by branching into either the myenteric plexus, the submucosal plexus, or mucosa. To determine whether activation of DRG subtypes from the colon is sufficient to drive pain responses, mouse optogenetic experiments were done using genetic labeling strategies and optogenetic probe placement in the distal colon. Activating colon innervating $CGRP^+$ neurons or the $Bmpr1b^+$ Aδ HTMRs evoked robust pupil dilation, movement, and vocalizations. Activation of the $Sstr2^+$ C-HTMRs led to an increase in pupil dilation and vocalizations but not movement. In contrast, activation of the $TrkB^+$ LTMRs did not lead to a noticeable behavioral response. Finally, we assessed the functional consequences of $Bmpr1b^+$ neuron ablation and subjecting control and ablated animals to balloon distension with or without dextran sulfate sodium (DSS) induced inflammation. Ablating $Bmpr1b^+$ sensory neurons in both DSS treated and non-treated animals led to a reduction in the pupil dilation response during balloon distension compared to DSS treated and non-treated control animals. Our findings suggest that the colon innervating $Bmpr1b^+$ Aδ-HTMRs mediate pain responses and contribute to mechanical hypersensitivity in the context of inflammation.

Disclosures: A. Abdelaziz: None. **R. Wolfson:** None. **S. Kushner:** None. **D. Ginty:** None.

Poster

PSTR140. Interoception

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR140.08/R1

Topic: D.02. Somatosensation – Pain

Support: NIH R01 DK120824
NIH U01 NS113873

Title: Functional characterization of chemo- and mechanosensitive afferents innervating distal colon and rectum

Authors: *N. DEIERLEIN[1], B. FENG[2], T. GUO[2], L. CHEN[2], Z. VAHEDI[2], G. ZHENG[2], S. FENG[2];
[1]Univ. of Connecticut Biomed. Engin. Dept., Storrs, CT; [2]Univ. of Connecticut, Storrs, CT

Abstract: The cardinal complaint from patients with irritable bowel syndrome (IBS) is abdominal and bowel pain, which arises from the neural encoding of sensory afferents innervating the large intestine. Accumulating clinical and preclinical evidence has indicated that afferent sensitization drives the prolonged visceral pain and hypersensitivity in IBS. Although mechanical distension of hollow visceral organs is the most reliable modality to evoke visceral pain perception, the altered chemical environment within the intestinal lumen, i.e., osmolarity and inflammatory mediators, likely contributes to afferent sensitization to mechanical stimuli. In this study, we implemented high-throughput optical recordings from dorsal root ganglion (DRG) neurons to systematically characterize the functional responses of afferents innervating the distal colon and rectum (colorectum) to chemical and mechanical stimuli. We systematically recorded GCaMP6f signals from a total of 829 individual afferents from male and female mice with a C57BL/6 background heterozygous for VGLUT2-Cre and GCaMP6f. We induced behavioral visceral hypersensitivity by intracolonic zymosan treatment (30 mg/mL, 3 consecutive days). We harvested in continuity the colorectum, pelvic and lumbar splanchnic nerve, and thoracolumbar (T12-L2) and lumbosacral (L5-S1) DRGs. The colorectum was cannulated to allow the delivery of four mechanical and chemical stimuli: graded colorectal distension (15, 30, 45, 60 mmHg, 5 sec steps), mucosal shear flow (25 mL/min), acid hypertonic solution (800 mOsm, pH 6.0), and inflammatory soup (5-HT, bradykinin, histamine, prostaglandin E2). The evoked GCaMP6f signals will be recorded using an upright fluorescent microscope with a high-speed, ultra-low-noise sCMOS camera (Andor Xyla-4.2P, 82% quantum efficiency). The 829 afferents were functionally divided into six classes based on their response profiles: mechanosensitive, chemosensitive, polymodal, mechanosensitized, chemosensitized, and silent afferents. More afferents were recorded from the zymosan-treated hypersensitive group than the saline-treated control group. Our preliminary analyses indicate a significant sex difference in the change in afferent proportions following zymosan treatment. The outcomes of the current study provide neurophysiological evidence of the sex differences in afferent neural encoding and sensitization, justifying future focused research to consider differential treatments between male and female IBS patients.

Disclosures: **N. Deierlein:** None. **B. Feng:** None. **T. Guo:** None. **L. Chen:** None. **Z. Vahedi:** None. **G. Zheng:** None. **S. Feng:** None.

Poster

PSTR140. Interoception

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR140.09/R2

Topic: D.02. Somatosensation – Pain

Support: NSF CAREER 1844762

Title: The biomechanical heterogeneity along the circumferential direction of the colorectum and its relevance to visceral nociception

Authors: *A. P. SECK, S. SIRI, K. HOSHINO, T. NADEEM, B. FENG;
Biomed. Engin., Univ. of Connecticut Biomed. Engin. Dept., Storrs, CT

Abstract: Nociceptive pain arising from the distal colon and rectum, known as the colorectum, is consistently induced by mechanical distention and stretch. This suggests the important role of tissue biomechanics in visceral nociception, as it determines the micromechanical environment around mechanosensitive afferent endings in the colorectal wall. Our prior study on colorectal inflation revealed that the colorectum ruptures uniformly along the mesentery, which is concentrated with extrinsic sensory afferent endings. In this study, we aim to systematically characterize the biomechanical properties of the colorectum along the circumferential direction, specifically between the mesenteric region and the adjacent antimesenteric regions. Conventional biaxial and uniaxial extension tests are inadequate for studying tissue-injurious levels of stretch due to significant stress concentration at the boundary fixtures. Therefore, we developed a new approach that involves stretching the tubular colorectum using two circumferential metal rods with a diameter of Φ1.2 mm. This approach significantly reduces stress concentration at the fixtures, enabling large strain deformation under tissue-injurious circumferential stretch. We harvested the distal 25 mm of the colorectum from C57BL/6 mice and transgenic mice heterozygous for VGLUT2-Cre and tdTomato genes (VGLUT2/tdT). A customized image tracking software was used to extract local strain profiles, and nonlinear imaging with second harmonic generation (SHG) was conducted to determine the collagen fiber content and orientation in the colorectum. Our results showed comparable strain levels between the mesenteric and antimesenteric regions in the colorectum when undergoing circumferential stretch equivalent to intraluminal distension of 150 mmHg. This level of stretch is significantly beyond the noxious range of 20 mmHg in mice. The collagen fiber density and orientations measured from SHG also showed no significant difference along the circumferential direction of the colorectum. Additionally, our SHG and confocal microscopy results identified a higher concentration of large blood vessels in the mesenteric region compared to the antimesenteric region. These findings suggest that the absence of apparent biomechanical heterogeneity between the mesenteric and antimesenteric regions occurs under both innocuous and noxious distension. The mechanical failure resulting from tissue-injurious inflation likely arises from the presence of large blood vessels in the mesenteric region, contributing to local stress concentration.

Disclosures: **A.P. Seck:** None. **S. Siri:** None. **K. Hoshino:** None. **T. Nadeem:** None. **B. Feng:** None.

Poster

PSTR140. Interoception

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR140.10/R3

Topic: D.02. Somatosensation – Pain

Support: NIH U01 NS113868
NIH T32 HL160529

Title: Decoding the role of genetically-distinct sensory nerves in controlling cardiopulmonary reflexes

Authors: *T. DARCEY, T. TAYLOR-CLARK;
Univ. of South Florida, Tampa, FL

Abstract: Activation of airway sensory nerves causes respiratory and autonomic reflexes. The majority of the airway sensory afferents are only sensitive to noxious stimuli, such as inflammation, infection, irritants, and pollutants. Activation of these nociceptive afferents evokes protective mechanisms such as apnea, cough, and bradycardia. These reflexes may contribute to disease morbidity when excessively or inappropriately activated. Airway nociceptive sensory nerves, which are largely projected from the vagal ganglia (nodose and jugular ganglion), are heterogeneous with respect to gene expression and neuroanatomy. Our objective is to characterize the reflexes evoked by activation of specific afferent subsets using chemogenetics. To selectively activate vagal afferent subpopulations *in vivo*, mice were exposed to nebulized selective stimuli such as capsaicin (transient receptor potential (TRP) vanilloid 1 (V1) agonist), allyl isothiocyanate (AITC, TRP ankyrin 1 (A1) agonist), and clozapine-N-oxide (CNO, selective agonist for the designer receptors exclusively activated by designer drugs (DREADD) stimulatory receptor hM3Dq). hM3Dq expression by ROSA26-loxP-STOP-loxP mice was selectively expressed in sensory subpopulations by crossing with cre recombinase expressing mice: TRPV1-cre (all nociceptors), Tac1-cre (peptidergic/jugular-originating nerves), and P2X2-cre (nodose-originating nerves). An intersectional approach using TRPV1-flp mice and the above cre mice and bilateral vagal injections of an AAV delivering dual recombinase-dependent DREADD constructs was used to express hM3Dq in afferent populations defined by two genes (e.g. TRPV1+ and P2X2+). ECGs were recorded via radiotelemetry, and respiration was measured via whole-body plethysmography. Capsaicin and AITC produced comparable levels of bradypnea, but AITC evoked greater bradycardia than capsaicin. Bradypnea also appeared at lower doses for both capsaicin and AITC compared to the higher doses required to evoke bradycardia. CNO administration to TRPV1-hM3Dq mice, P2X2-hM3Dq mice, and Tac1-hM3Dq mice evoked bradypnea and bradycardia. This suggests that both nodose afferents and jugular afferents evoke similar reflexes. Nevertheless, CNO exposure to mice with hM3Dq

expressed only in the nodose nociceptor population (TRPV1+/P2X2+) population had little effect on resting heart rate and furthermore inhibited subsequent AITC-evoked bradycardia. Thus, our data suggests that functionally-distinct nociceptive subpopulations may differentially regulate cardiopulmonary reflexes.

Disclosures: **T. Darcey:** None. **T. Taylor-Clark:** None.

Poster

PSTR140. Interoception

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR140.11/R4

Topic: D.02. Somatosensation – Pain

Support: NIH Common Fund SPARC OT2

Title: Generation of Novel Cre and Flp Dependent Dual Reporter Strain to Intersectionally Label Vagal Afferent Nerve C-fiber Subtypes

Authors: *S.-H. KIM, M. PATIL, J. FIALLO, A. GUERRERO, S. HADLEY, T. TAYLOR-CLARK;
Univ. of South Florida, Tampa, FL

Abstract: Bronchopulmonary reflexes are controlled by vagal afferent nerves. The vagal afferent nerves, which are comprised of jugular (neural crest origin) and nodose (placodal origin), exert distinct airway reflexes. Capsaicin receptor TRPV1 is selectively expressed on vagal nociceptors that sense noxious stimuli. Tac1 encodes tachykinin1 and is mainly expressed in the jugular ganglia. Purinergic receptor P2X2 expression is exclusively in nodose ganglia. TRPV1 expressing C-fibers are present in both jugular and nodose ganglia, so to visualize nodose and jugular nociceptors we used an intersectional genetics approach. First, we generated a transgenic mouse model with dual recombinase responsive allele using Cre-Lox and Flp-FRT systems. With the ROSA26-RC::FLTG allele, cells with only Flp produce tdTomato expression while cells with both Flp and Cre produce GFP expression. We generated TRPV1^{Flp}:Tac1^{Cre}:ROSA and TRPV1^{Flp}:P2X2^{Cre}:ROSA strains which we characterized. Next, we unilaterally injected AAV9-Ef1a-C$_{ON}$F$_{ON}$-mCherry into the vagal ganglia, which requires the presence of both Cre and Flp recombinases for mCherry expression in infected cells, into TRPV1^{Flp}:Tac1^{Cre} and TRPV1^{Flp}:P2X2^{Cre} strains to selectively target jugular nociceptors and nodose nociceptors, respectively. In TRPV1^{Flp}:Tac1^{Cre}:ROSA, the majority of TRPV1^{+}/Tac1^{-} cells were only in nodose (14.01%) and TRPV1^{+}/Tac1^{+}neurons were mostly in jugular (16.38%). In TRPV1^{Flp}:P2X2^{Cre}:ROSA, TRPV1^{+}/P2X2^{+} neurons were only observed in nodose (15.10%) while TRPV1^{+}/P2X2^{-} neurons were observed in both nodose (4.35%) and jugular ganglia (5.72%). With vagal injection of AAV9-Ef1a-C$_{ON}$F$_{ON}$-mCherry, mCherry expressing TRPV1^{+}/Tac1^{+} cells were found only in jugular in TRPV1^{Flp}:Tac1^{Cre}, while TRPV1^{+}/P2X2^{+} cells were found only in nodose in TRPV1^{Flp}:P2X2^{Cre} strain. In the brainstem, reporter expression was

mainly in the nucleus of solitary tract (nTS) in both strains, but only $TRPV1^{+}/Tac1^{+}$ innervation was observed in the paratrigeminal nucleus (Pa5) in $TRPV1^{Flp}$:$Tac1^{Cre}$:ROSA as well as in $TRPV1^{Flp}$:$Tac1^{Cre}$ strain injected with AAV-Ef1a-C$_{ON}$F$_{ON}$-mCherry. In the lung, most $TRPV1^{+}/Tac1^{+}$ fibers were found near the epithelial layer of bronchi and bronchioles, whereas $TRPV1^{+}/P2X2^{+}$ fibers often projected away from the conducting airways into the alveolar tissue. Taken together, we have generated cell type specific-dual reporter mouse models using a combinatorial/intersectional Cre and Flp strategy to successfully identify distinct nociceptive subtypes of afferent nerves in vagal ganglia, brainstem, and lung.

Disclosures: S. Kim: None. **M. Patil:** None. **J. Fiallo:** None. **A. Guerrero:** None. **S. Hadley:** None. **T. Taylor-Clark:** None.

Poster

PSTR140. Interoception

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR140.12/R5

Topic: D.02. Somatosensation – Pain

Title: Using tissue clearing and light-sheet fluorescence microscopy for the three-dimensional analysis of sensory and sympathetic nerve endings that innervate bone

Authors: *J. THAI, J.-P. FULLER-JACKSON, J. IVANUSIC;
Anat. and Physiol., Univ. of Melbourne, Melbourne, Australia

Abstract: Bone and bone marrow are richly innervated by sensory and sympathetic nerve fibers. However, characterisation of the morphology, molecular phenotype and distribution of nerves that innervate hard tissue has so far been limited to thin histological sections that does not adequately capture dispersed neuronal projections due to the loss of structural information during 3D reconstruction. In this study, we modified the iDISCO/iDISCO+ clearing protocol to image high-resolution 3D neuronal structures in whole femurs collected from perfused C57BL/6 mice (n=11). Nerve fibers and their endings were immunolabelled with antibodies directed against protein gene product 9.5 (pan-neuronal marker; PGP9.5), calcitonin gene-related peptide (peptidergic nociceptor marker; CGRP), or tyrosine hydroxylase (sympathetic neuron marker; TH). Volume imaging was performed using light-sheet fluorescence microscopy. PGP9.5+ nerve bundles entered the marrow cavity via nutrient foramina and branched extensively into the trochanter, the marrow cavity, and the epiphyses. They often followed blood vessels through the marrow cavity, and terminated with a corkscrew morphology around blood vessels, or as free endings away from blood vessels. The free endings were mostly simple, some with *en passant* varicosities along their length and some without. As they ran down the femur, they gave branches that projected laterally to terminate near the endosteum, or left the marrow cavity through Volkmann's and Haversian canals in the cortical bone. Innervation density decreased lower in the femur, and was least in the lower third of the diaphysis and distal metaphysis. CGRP+ nerve fibers and endings often followed blood vessels, but did not have the corkscrew

morphology observed for some of the PGP9.5+ fibers. They typically had numerous *en passant* type varicosities along their length, and most often terminated away from blood vessels as free endings in the marrow cavity or near the endosteum. In contrast, TH+ nerve fibers and endings typically spiralled around blood vessels, and less frequently terminated as free endings away from blood vessels. Notably, we observed distinct clusters of complex endings associated with the termination of a single axon in the lower diaphysis. Those that were CGRP+ had a different morphology to those that were TH+. Our findings show evidence of differential distribution patterns and morphology for sensory and sympathetic neurons within bone. Mapping the distribution of these endings in different bone marrow niches may be important in further elucidating their roles during homeostasis, and in injury or disease.

Disclosures: J. Thai: None. **J. Fuller-Jackson:** None. **J. Ivanusic:** None.

Poster

PSTR140. Interoception

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR140.13/R6

Topic: D.02. Somatosensation – Pain

Title: Towards a further understanding of bone pain: insights from mechanical and optical stimulation of bone afferent neurons in mice

Authors: M. MORGAN[1], H. LEE[1], A. MOE[3], J. THAI[1], ***J. IVANUSIC**[2];
[2]Anat. and Physiol., [1]Univ. of Melbourne, Melbourne, Australia; [3]Monash Univ., Melbourne, Australia

Abstract: Our laboratory has recently developed a highly novel and innovative, *in vivo* electrophysiological bone-nerve preparation that allows us to record the activity and sensitivity of peripheral sensory neurons that innervate the rat tibia. We have exploited this preparation to identify how noxious mechanical stimuli and/or known algesic substances contribute to bone pain, both in naïve animals and in an animal model of OA. This preparation has provided unprecedented insight into mechanisms that generate and maintain bone pain, but the typical experimental design is limited to using pharmacological manipulation of specific ion channels or receptors that we are interested in. Here we aimed to develop and characterize an electrophysiological bone-nerve preparation that allows us to record sensory neurons that innervate bone in mice, so we can take advantage of transgenic approaches in the future. Experiments were performed in isoflurane anesthetized C57BL/6 mice. A fine branch of the tibial nerve that innervates the marrow cavity was identified and placed over a platinum hook electrode for extracellular recording. A reference electrode was implanted into a nearby muscle. Whole-nerve electrical activity was amplified (1000×) and filtered (high pass 100 Hz, low pass 3 kHz), sampled at 20 kHz and stored to PC. Mechanical stimulation was delivered by injecting saline into the marrow cavity, through a canula, to increase intraosseous pressure. Capsaicin (2 μM) was delivered into the marrow cavity using the same canula. In most recordings, 2-3 single

mechanically sensitive units could be discriminated by their spike amplitude and shape. We report that murine bone afferent neurons respond only to high threshold noxious mechanical stimulation, code for the intensity of mechanical stimulation, can be sensitized by capsaicin but not by saline, and do not suffer stimulus-evoked fatigue when using 10 min interstimulus intervals. To achieve and confirm optical stimulation of bone afferent neurons in mice, we made electrophysiological recordings of the same nerve in response to application of 473 nm blue light (1 Hz, 0.25-0.5 ms, 5-63 $mWmm^{-2}$) to the tibial marrow cavity in *Wnt1-Cre;loxP-ChR2* mice. We report compound action potentials with conduction velocities in the Aδ and C fiber range with optical stimulation of the marrow cavity, but no response with optical stimulation of the periosteum. These new approaches to recording the activity and sensitivity of bone afferent neurons will allow us to take advantage of transgenic tools to further our understanding of mechanisms that generate and maintain bone pain in the future.

Disclosures: M. Morgan: None. **H. Lee:** None. **A. Moe:** None. **J. Thai:** None. **J. Ivanusic:** None.

Poster

PSTR140. Interoception

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR140.14/Web Only

Topic: D.02. Somatosensation – Pain

Support: HL152160
HL126769

Title: Transient Receptor Potential Vanilloid 1 (TRPV1) Channels in Lumbar Dorsal Root Ganglia are downregulated in a Rodent Model of Chronic Peripheral Arterial Disease

Authors: *N. KARPUK[1], H. WANG[2];
[2]UNMC Address, [1]Univ. of Nebraska Med. Ctr., Omaha, NE

Abstract: Introduction- Peripheral arterial disease (PAD) is a manifestation of systemic atherosclerosis affecting approximately 8 to 12 million people in the United States. The signs and symptoms of PAD, including claudication, resting pain, and tissue loss, are consequences of both skeletal myopathy and skeletal muscle sensory dysfunction. However, the molecular mechanisms underlying muscle sensory dysfunction in PAD remains unclear. Here, we hypothesized that Transient Receptor Potential Vanilloid 1 (TRPV1) channels are upregulated in lumbar dorsal root ganglia (DRG) and play a critical role in mediating muscle sensory dysfunction in rats and mice with chronic hindlimb ischemia. **Methods**-We developed a novel endovascular femoral artery occlusion rodent (rat and mouse) model in which a catheter is inserted into the femoral artery to the aortic bifurcation. To validate our model, we have assessed hindlimb perfusion using Scanning Laser Doppler in occluded rat/mice and sham-surgery. We first examined time-dependent TRPV1 protein expression in ipsilateral L4/L5 DRGs of

endovascular femoral arterial occlusion-induced PAD rats. We also performed *ex vivo* calcium imaging in response to bath application of capsaicin (0.5 μM) in lumbar DRGs from TRPV1-specific GCAMP6 reporter mice that were sham-operated and compared to endovascular femoral arterial occlusion. Statistical comparisons were performed by one-way ANOVA or Mann-Whitney test. **Results**- The endovascular catheter greatly reduced blood flow in both rats and mice to the hind paw up to 6 weeks. Recovery of flow started at 6 weeks but remained significantly reduced. We found that TRPV1 receptors were initially elevated in the acute PAD phase (e.g. 1 week post occlusion) but significantly decreased in the chronic PAD phase (i.e. 4 weeks post occlusion), suggesting a dynamic change of TRPV1 receptors during acute and chronic phases of PAD. Similarly, we also found that capsaicin-evoked calcium influx in lumbar DRG neurons were significantly reduced in PAD mice at 4 weeks post endovascular femoral arterial occlusion compared to sham mice (Sham vs. PAD: 0.61±0.02 vs. 0.32±0.01 ΔF/F0, Mean ± SE, $P<0.0001$, n=216 lumbar DRG neurons in sham and n=341 lumbar DRG neurons in PAD). **Conclusion**- Our data suggest that in chronic PAD, TRPV1 protein expression as well as its channel activity are downregulated in lumbar DRG neurons from chronic PAD animals. These data suggest that TRPV1 receptors in lumbar DRGs are unlikely to mediate muscle sensory dysfunction such as intermittent claudication in chronic PAD.

Disclosures: **N. Karpuk:** None. **H. Wang:** None.

Poster

PSTR140. Interoception

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR140.15/R7

Topic: F.08. Food and Water Intake and Energy Balance

Support: NIH Grant R01 AT011653
NIH Grant R03 TR003313
NIH Grant R01 DK092246
NIH Grant P30 DK020541

Title: Identification of vagal sensory neurons innervating the liver in mice

Authors: *W.-H. CHO, J. HWANG, Y.-H. JO;
Albert Einstein Col. of Med., New York, NY

Abstract: IDENTIFICATION OF VAGAL SENSORY NEURONS INNERVATING THE LIVER IN MICE.Woo-Hyun Cho[1,2*], Jiyeon Hwang[1,2] , , and Young-Hwan Jo[1,2,31]The Fleischer Institute for Diabetes and Metabolism, [2]Division of Endocrinology, Department of Medicine and [3]Department of Molecular Pharmacology, Albert Einstein College of Medicine, NY, USAThe neural connection between the brain and the liver consists of branches of the sympathetic and parasympathetic autonomic nervous systems. In addition, vagal sensory neurons in the nodose ganglion innervate the liver. However, it remains unknown whether vagal sensory neurons

innervating the liver can sense and respond to interoceptive signals such as nutrients and other metabolic signals and transmit these interoceptive signals to the brain. This study aimed to determine the cellular identity of vagal sensory neurons innervating the liver.To identify vagal sensory neurons innervating the liver, we used the advillin (Avil)^{CreERT2} transgenic strain expressing a tamoxifen-inducible Cre recombinase directed by mouse advillin promoter. Following laparotomy, we injected a total volume of 20 μl of a Cre-dependent retrograde viral tracer AAVrg-FLEX-tdTomato (titer, 1.3 X 10^{13} pfu/ml) into the left and medial lobes of the livers of Avil^{CreERT2} mice. In these experimental conditions, the viruses were taken up by axon terminals and transported to the cell body, permitting to induce Cre-mediated tdTomato expression exclusively in the vagal sensory neurons projecting to the liver. Nodose ganglion sections were stained with anti-tdTomato and anti-Avil antibodies. We found that a small subset of Avil-positive sensory neurons in the nodose ganglion expressed tdTomato. In addition, we found that Avil-positive sensory nerves in the liver were also labeled with an anti-calcitonin gene-related peptide (CGRP) antibody. As each vagal sensory neuron has a central brainstem terminal, we further examined if the brainstem receives interoceptive information from liver-innervating vagal sensory neurons. Our immunostaining revealed that the nucleus tractus solitarius (NTS) received synaptic input from vagal sensory neurons innervating the liver and that a subset of cholinergic neurons in the dorsal motor nucleus of the vagus (DMV) were also innervated by vagal sensory neurons innervating the liver. Hence, Our preliminary results suggested that a subset of chemosensitive Avil-, and CGRP-positive neurons were liver-projecting vagal sensory neurons. These vagal sensory neurons may play a role in regulating metabolic homeostasis in mice.

Disclosures: W. Cho: None. **J. Hwang:** None. **Y. Jo:** None.

Poster

PSTR140. Interoception

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR140.16/R8

Topic: F.08. Food and Water Intake and Energy Balance

Support: NIH Grant R01 AT011653
NIH Grant R03 TR003313
NIH Grant R01 DK092246
NIH Grant P30 DK020541

Title: Identification of parasympathetic cholinergic neurons innervating the liver in mice

Authors: *J. HWANG, W.-H. CHO, Y.-H. JO;
Albert Einstein Col. of Med., Bronx, NY

Abstract: The dorsal motor nucleus of the vagus (DMV) contains parasympathetic cholinergic neurons. These cholinergic neurons innervate various internal organs, including the liver. Prior

neuroanatomical studies demonstrate that a subset of DMV cholinergic neurons project to the liver. Our recent study further shows that hepatocytes receive direct DMV cholinergic input and express the muscarinic acetylcholine receptor genes. Optogenetic inhibition of vagal cholinergic neurons innervating the liver elevates hepatic glucose output, suggesting that the hepatic cholinergic system contributes to hepatic metabolism. In contrast to these findings, recent 3D imaging studies of cleared tissues show a lack of parasympathetic cholinergic nerves in the mouse liver. This study aimed to re-evaluate the cholinergic system in the liver to better understand how the parasympathetic nervous system of the liver controls metabolic homeostasis. In this study, we injected a Cre-dependent viral tracer (adeno-associated virus (AAV).PHP.eB-FLEX-tdTomato) into the livers of choline acetyltransferase (ChAT)-IRES-Cre ($ChAT^{Cre}$) mice. Double immunostaining was carried out with anti-ChAT and anti-tdTomato antibodies. Approximately 10 % of DMV cholinergic neurons expressed tdTomato and all tdTomato-positive cells were ChAT-expressing cells (n = 72 out of 72 neurons from 3 mice). We then examined if the liver receives synaptic input from DMV cholinergic neurons by injecting an anterograde AAV1 encoding a Cre-inducible tdTomato and a synaptophysin-GFP (sypGFP) fusion protein that are separated by the sequence *of a* T2A *self-cleaving* peptide *(*AAV1-FLEX-tdTomato-T2A-sypGFP) to the DMV of $ChAT^{Cre}$ mice. Immunostaining revealed that DMV cholinergic neurons were positive for tdTomato and that numerous sypGFP-positive puncta were found in hepatocytes and bile duct epithelial cells cholangiocytes. Immunostaining with an antibody against the presynaptic cholinergic marker vesicular acetylcholine transporter (vAChT) revealed that puncta of vAChT-positive nerve terminals were detected throughout the liver parenchyma. Western blot analysis of liver tissue homogenates revealed the presence of the muscarinic ACh receptor type 1, 2, 4, and 5. Hence, our results strongly support the presence of the cholinergic system in the mouse liver.

Disclosures: **J. Hwang:** None. **W. Cho:** None. **Y. Jo:** None.

Poster

PSTR140. Interoception

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR140.17/S1

Topic: D.02. Somatosensation – Pain

Support: FWO G0E0520N
ERC 945539
KU Leuven C14/21/111

Title: Whole-brain functional mapping of time-varying heart rate variability in the awake macaque

Authors: *T. POPPA[1,2,3], R. E. HARTIG[5], D. MANTINI[1,3,4], W. VANDUFFEL[1,6,7,3,2], H. C. EVRARD[8,9,10,5];
[2]Lab. for Neuro- and Psychophysiology, [3]Leuven Brain Inst., [4]Movement Control and

Neuroplasticity Res. Group, [1]Katholieke Univ. Leuven, Leuven, Belgium; [5]Nathan Kline Inst. for Psychiatric Res., Orangeburg, NY; [6]A. A. Martinos Ctr. for Biomed. Imaging, MGH, Charleston, MA; [7]Dept. of Radiology, Harvard Med. Sch., Boston, MA; [8]Max Planck Inst. For Biol. Cybernetics, Tübingen, Germany; [9]Intl. Ctr. for Primate Brain Research, Chinese Acad. of Sci., Shanghai, China; [10]Werner Reichardt Ctr. for Integrative Neuroscience, Univ. of Tübingen, Tübingen, Germany

Abstract: Human fMRI studies suggest that the autonomic nervous system (ANS) interacts with a central autonomic network (CAN) that is anchored in the bilateral anterior insular, mid-cingulate and inferior parietal cortices. CAN integrity bears relevance to ANS dysregulation in neurological and psychiatric disorders. Establishing the existence of a homolog network in the macaque could greatly facilitate systems-level investigations into causal CAN-ANS dynamics. Here, we recorded electrocardiography (ECG) and respiration in an awake, trained rhesus macaque during contrast-agent enhanced, whole-brain, high-resolution fMRI with implanted phased-array coils (0.6 mm isotropic voxels at 3T). Time-resolved estimates of parasympathetically-mediated, high frequency heart rate variability (hfHRV) were extracted from ECG using a Maximal Overlap Discrete Wavelet Packet Transform, subsequently down-sampled to TR resolution (3 sec), normalized, and convolved with a hemodynamic response function. The convolved hfHRV time series were regressed on the fMRI time series, while accounting for respiratory and cardiac noise via RETROICOR modeling. HfHRV was robustly anti-correlated with a network that prominently included the bilateral dorsal anterior insula (dAI), posterior and mid-cingulate cortices, precentral gyrus, and central sulcus. Subcortically, the network included the bilateral thalamus and putamen ($p < 0.05$, FWE-corrected). To test whether the configuration of CAN connectivity with the left and right dAI depended on fluctuations in cardiovagal state, we used the dAI time series and HRV time series to perform a physio-physiological interaction analysis. This revealed that functional connectivity of the left (but not right) dAI with the posterior, middle and anterior cingulate cortices, and the bilateral ventral dysgranular insula depends on cardiovagal state ($p < 0.001$), concordant with a previously proposed leftward parasympathetic dominance model. The striking similarity of human and macaque CANs that (anti)correlate with time-resolved estimates of hfHRV suggests substantial evolutionary conservation. The identified CAN has regional specificity characterized by contributions from cingulo-operculo-insular and sensorimotor systems. This pattern of coactivation may be consistent with original conceptualizations of the salience network as serving a domain general role in coordinating autonomic, behavioral, and cognitive responses to homeostatically relevant internal and external events.

Disclosures: T. Poppa: None. **R.E. Hartig:** None. **D. Mantini:** None. **W. Vanduffel:** None. **H.C. Evrard:** None.

Poster

PSTR140. Interoception

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR140.18/S2

Topic: D.02. Somatosensation – Pain

Support: FWO G0E0520N
KU Leuven C14/21/111
Shanghai Municipal Science and Technology Major Project (Grant No. 2019SHZDZX02)

Title: Thermosensory activation of insular cortex in the awake macaque

Authors: *H. C. EVRARD[1,2,3,4], T. POPPA[5,6], R. E. HARTIG[2], A. MOURAUX[7], W. VANDUFFEL[8,6,9];
[1]Intl. Ctr. for Primate Brain Res., Shanghai, China; [2]Nathan Kline Inst. for Psychiatric Res., Orangeburg, NY; [3]Werner Reichardt Ctr. for Integrative Neuroscience, Univ. of Tübingen, Tübingen, Germany; [4]Max Planck Inst. for Biol. Cybernetics, Tübingen, Germany; [5]Lab. for Neuro- and Psychophysiology, KU Leuven, Leuven, Belgium; [6]Leuven Brain Inst., Leuven, Belgium; [7]Inst. of Neurosci. (IONS), Univ. Catholique De Louvain, Brussels, Belgium; [8]A.A. Martinos Ctr. for Biomed. Imaging, MGH, Charlestown, MA; [9]Radiology, Harvard Med. Sch., Charlestown, MA

Abstract: The posterior dorsal granular fundus of the insular cortex (IDFP) is the terminus of a phylogenetically recent spino-thalamo-cortical pathway, representing various interoceptive modalities, including thermoception. Prior tract-tracing and electrophysiological recordings in the anesthetized macaque, and neuroimaging in humans, indicated that thermoception is encoded along a posterior-to-anterior sacral-to-trigeminal somatotopy, with the face being represented in the most anterior portion of IDFP. Yet, direct evidence for the occurrence of a thermoceptive representation of trigeminal afferent in the awake macaque monkey is still needed. Here, we used an infrared Nd:YAP laser adapted for MR-compatibility to deliver 20 ms pulses of radiant heat over 0.9 mm diameter targets, along the maxillary division of the face, during contrast-agent enhanced, whole-brain, high-resolution fMRI (0.6 mm isotropic voxels at 3T) in an awake rhesus monkey implanted with 8-channel receiver coils. Low and high laser intensities were set to activate trigeminal C-/Aδ primary afferent fibers with temperatures reaching ~43°C and ~49°C, respectively (mean interstimulus interval = 21 sec). A GLM contrasting high intensity vs. null revealed activation predominantly in the anterior portion of IDFP, as well as in cortical areas 3a and 24, the posterior portion of the ventromedial thalamic nucleus, the periaqueductal gray, and the principal trigeminal nucleus, contralateral to the side of stimulation (voxel-wise, $p<0.001$). Additional activation occurred in the anterior insula, medial prefrontal area 32, lateral orbitofrontal area 11, and visual areas (V6), bilaterally. Split-half conjunction analysis of the high vs. null contrast further highlighted contralateral IDFP and bilateral anterior insula. Similar activations occurred in IDFP at more liberal thresholds when contrasting low intensity vs. null, consistent with prior evidence that the insula encodes stimulus intensity. Finally, epoched pupil diameter time-courses differentiated the conditions (cluster permutation, $p<0.05$). These results support the preponderance of a trigemino-thalamo-insular pathway for thermoception (including thermonociception) in primates with a localization of a face representation in anterior IDFP. Future analyses will test whether arousal (pupil dilation and heart rate variations) correlates with the thermosensory activation in bilateral anterior insula and extrastriate visual area V6, which may elucidate the contributions of salience and attention, respectively, in the processing of thermoception in primates.

Disclosures: **H.C. Evrard:** None. **T. Poppa:** None. **R.E. Hartig:** None. **A. Mouraux:** None. **W. Vanduffel:** None.

Poster

PSTR140. Interoception

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR140.19/S3

Topic: D.02. Somatosensation – Pain

Support: Max Planck Society
Shanghai Municipal Science and Technology Major Project (Grant No. 2019SHZDZX02)

Title: Von Economo neuron projections to brainstem autonomic and interoceptive nuclei in the macaque monkey

Authors: *G. CHAVEZ MARCHETTA[1,2], F. M. HORN[1,2], T. O. SALEH[1,2], H. KENNEDY[3,4], H. C. EVRARD[1,2,5,6];
[1]Max Planck Inst. for Biol. Cybernetics, Tubingen, Germany; [2]Werner Reichardt Ctr. For Integrative Neurosci., Tubingen, Germany; [3]Univ. Lyon, Univ. Claude Bernard Lyon 1, Inserm, Stem Cell and Brain Res. Inst. U1208, Bron, France; [4]Inst. of Neuroscience, State Key Lab. of Neuroscience, Chinese Acad. of Sci., Shanghai, China; [5]Intl. Ctr. for Primate Brain Research, Chinese Acad. of Sci., Shanghai, China; [6]Nathan Kline Inst. for Psychiatric Res., Orangeburg, NY

Abstract: The von Economo (VEN) and fork (FN) neurons are unique neuronal morphotypes found predominantly in the anterior insular and cingulate cortices (AIC and ACC) in primates and in a few other mammals. We previously showed that the VEN, FN and a relatively small group of local pyramidal neurons (PN) project to the periaqueductal gray (PAG) and parabrachial nucleus (PBN), supporting the idea of interoceptive prediction errors mitigation through sensory gating and autonomic control. Prior studies in macaques concluded that AIC and ACC do not project to further caudal brainstem nuclei, hereby limiting their top-down influence. Here, we examined the distribution of axon terminals with injections of anterograde tracers in AIC, as well as the distribution of cortical somata with injections of retrograde tracers in the lateral amygdala (LA), PAG, PBN, locus coeruleus (LC), caudal brainstem solitary (Sol), dorsal motor vagus (DMV) and retroambigus (RAmb) nuclei, and cervical spinal cord (C5-7). Anterograde labeling occurred in all the diencephalic, midbrain and brainstem nuclei. Sparse labeling occurred in the corticospinal pyramidal tract and in lamina 1 in the 1st cervical segment. Retrograde labeling of all three neuronal morphotypes occurred in layer 5 of AIC and ACC with injections in LA, PAG, PBN, and lower brainstem, but not in C5-7. The ratio of retrogradely labeled VENs over the total number of labeled neurons in layer 5 in AIC was significantly higher (PBN, 28%; PAG, 17%; LA, 21-26.3%; LC, 13.3%; Sol/RAmb, 10.8%) than the overall VEN population number in adjacent Nissl-stained sections (3%). To further investigate the

corticospinal projections, we examined the labeling produced with injections of retrograde tracers in C5-7 in adults vs prenatal E95 and E105 macaques. VEN, FN, and PN were labeled in AIC at E95 and E105 (E95 more than E105) but not in adults.
This study shows that VEN and FN belong to a population of projecting neurons that can potentially influence a much broader set of subcortical regions than previously assumed. Most, if not all, brain regions involved in interoceptive integration and autonomic regulation receive AIC and ACC projections. The added functional value of the VEN and FN to these projections could hypothetically resemble that of the Betz cells in motor control. The corticospinal projections require further investigation, with longer post-injection survival and more caudal injection sites, to test whether only specific spinal segments receive VEN projections (e.g., sympathetic thoracolumbar columns) or whether all corticospinal projections are indeed pruned during development. The latter would constitute a remarkable finding.

Disclosures: G. Chavez Marchetta: None. **F.M. Horn:** None. **T.O. Saleh:** None. **H. Kennedy:** None. **H.C. Evrard:** None.

Poster

PSTR141. Molecular and Cellular Pain Models

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR141.01/S4

Topic: D.02. Somatosensation – Pain

Support: NIHR01DE029074

Title: Upregulation of Fos and peptide markers in the lumbosacral, but not thoracolumbar, spinal cord contributes to visceral hypersensitivity in a rat model of Chronic Overlapping Pain Conditions.

Authors: *H. MEKONEN, E. KO, S. PANDYA, R. TRAUB;
Univ. of Maryalnd, Baltimore, MD

Abstract: Nociplastic chronic pain conditions, including temporomandibular disorder (TMD) and irritable bowel syndrome (IBS) are collectively known as Chronic Overlapping Pain Conditions (COPCs). They have greater prevalence in women, are exacerbated by stress and co-occur in greater than 50% of patients. However, underlying causes of individual conditions and the mechanisms contributing to their comorbidity are unclear. In a longitudinal study of Comorbid Pain Hypersensitivity (CPH) in rats (masseter inflammation + 4 days restraint stress inducing CPH), visceral hypersensitivity was assessed by measuring referred pain (increased mechanosensitivity of the lower back to von Frey stimulation), and analyzing expression of c-Fos, substance P and CGRP in the T12-L2 (TL) and L6-S1 (LS) spinal cord. These markers play a crucial role in pain studies as they contribute to our understanding of the neurobiological mechanisms underlying pain perception, transmission, modulation, and neurological inflammation. Referred pain was assessed in male and female naïve and CPH rats at 1, 4 and 7

weeks. CPH increased referred pain in females that persisted at least 7 weeks. In contrast, CPH increased referred pain in males that resolved by 4 weeks (2 way ANOVA, time p=0.0001; treatment p<0.01). In preliminary histochemical studies, male (n=4) and female (n=3) data were pooled. Preliminary results suggest a tendency towards an increase in Fos expression in the LS spinal cord at 1 week in CPH rats compared to naïve (p=0.09). There was no increase in the TL spinal cord. SP and CGRP expression in the LS and TL spinal cord were examined by fluorescence densitometry. There was an increase in CGRP density in the LS spinal cord in CPH rats at 1 week compared to naïve (p=0.0339). There was a tendency for an increase in SP density in the LS spinal cord (p=0.0806). There was no difference in SP or CGRP expression in the TL spinal cord. In the current study, visceral hypersensitivity was induced without single housing animals for months following EMG electrode surgery. Furthermore, since colorectal distention was not used, the referred visceral hypersensitivity is likely an indication of spontaneous visceral pain. Interestingly, distension-evoked visceral pain and spontaneous visceral pain had a similar time course, yet spinal processing might be different. Our preliminary histochemical data suggests spontaneous pain in this model might be independent of increased activity in the TL spinal cord while inflammatory or distention-evoked pain upregulates activity in the TL spinal cord. These data suggest that nociplastic visceral pain might activate different circuitry than inflammatory visceral pain

Disclosures: H. Mekonen: None. **E. Ko:** None. **S. Pandya:** None. **R. Traub:** None.

Poster

PSTR141. Molecular and Cellular Pain Models

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR141.02/S5

Topic: D.02. Somatosensation – Pain

Support: National Natural Science Foundation of China (81971049 and 81671097)
NIH R01 grants (DE029074, DE029946, and NS091296)

Title: Chronic mild stress induces wide-spreading hyperalgesia: the role of CCK_1 receptors in the spinal cord

Authors: J.-H. LI[1], Y. GUO[1], F. CHEN[2], T.-Y. LIU[1], R. J. TRAUB[3], F. WEI[3], ***D.-Y. CAO**[3,1]; [1]Xi'an Jiaotong Univ. Col. of Stomatology, Xi'an, China; [2]Shaanxi Univ. of Traditional Chinese Medicine, the Second affiliated Hosp., Xianyang, Shaanxi, China; [3]Univ. of Maryland Sch. of Dent., Baltimore, MD

Abstract: Purpose: Chronic primary pain (CPP) occurs in the absence of structural or tissue injury and includes some comorbid pain conditions such as the temporomandibular disorders (TMD), fibromyalgia syndrome (FMS) and irritable bowel syndrome (IBS). CPP is commonly considered a consequence relating to chronic stress in prevalence of women. However, their relationship remains poorly understand. The aim of this study is to develop a new animal model

to see whether chronic stress induces wide-spreading hyperalgesia and investigate central mechanisms underlying CPP. **Methods:** The 21 day stress was performed in adult female C57BL/6 mice with procedures including forced swim stress (FSS), water avoidance stress (WAS), and restraint stress (RS). The nocifensive responses were conducted before, during, and after chronic stress. The anxiety- and depression-like behaviors were tested during and after the stress. To identify the involvement of central mechanisms, we also examined whether spinal CCK_1 receptors were involved in the development of somatic hyperalgesia induced by chronic stress via intrathecal injection of CCK_1 receptor antagonist CR-1505 for five consecutive days after the stress. **Results:** The thermal withdraw latency of the unilateral hindpaw and the mechanical withdrawal thresholds of hindpaw, masseter muscle, upper back, thigh and abdomen were significantly decreased in the stress group compared to the control animals. The level of plasma corticosterone was significantly higher in the stress group than that in the control group. The entries and time spent in the center zone of the open field, and the percentages of entries and time spent in the open arms of the elevated plus maze (EPM) were significantly decreased in stress-treated animals. The immobility time during FSS significantly increased with the increase of days of stress compared to the first day of stress procedure. The sucrose consumption was significantly reduced after stress. The expression of CCK_1 receptors significantly increased in L4-L5 spinal dorsal horn of stress mice. Intrathecal injection of CCK_1 receptor antagonist attenuated stress-induced wide-spreading hyperalgesia over the body. **Conclusions:** Our results indicate that chronic mild stress produces a long-lasting and central CCK-dependent wide-spreading hyperalgesia in female mice, along with anxiety- and depression-like behaviors, providing a new animal model of CPP for investigating underlying mechanisms of comorbid pain conditions and a potential target for functional pain relief.

Disclosures: J. Li: None. **Y. Guo:** None. **F. Chen:** None. **T. Liu:** None. **R.J. Traub:** None. **F. Wei:** None. **D. Cao:** A. Employment/Salary (full or part-time); Xi'an Jiaotong University College of Stomatology.

Poster

PSTR141. Molecular and Cellular Pain Models

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR141.03/S7

Topic: D.02. Somatosensation – Pain

Title: Large scale generation of iPSCs derived human sensory neurons for CIPN and pain research

Authors: *A. FATHI[1], L. HARMS[1], D. MAJEWSKI[1], M. DONEGAN[1], K. TOMOTOSHI[1], S. HILCOVE[2], S. SCHACHTELE[3], J. LIU[1];
[1]R&D, [2]Fujifilm Cell. Dynamics Inc., Madison, WI; [3]Fujifilm Cell. Dynamics Inc., Minneapolis, MN

Abstract: Sensory neurons (SN) of the peripheral somatic nervous system respond to variety of impulses from sensory organs including touch, position in space (proprioception), temperature, and pain. An accessible and consistent source of human sensory neurons is of great value for advancing pain research and for the development and screening of next-generation chemotherapy drugs that minimize chemotherapy-induced peripheral neuropathy (CIPN), hematologic toxicity, and other side effects that alter quality of life for cancer patients. Methods for differentiation of human pluripotent cells into biologically-relevant human sensory neurons have experienced significant challenges when producing these neurons consistently at large scale. Here we characterize human induced pluripotent stem cell (iPSC)-derived sensory neurons produced via directed differentiation at large scale. These iPSC-derived SN have high purity (>80% BRN3A+/UCHL1+) and express hallmark nociceptive channels (ie. Nav1.7 and Nav1.8) and receptors (ie.TRPV1 and P2RX). We characterized sensory function of these neurons using electrophysiology and calcium imaging. Notably, we confirmed the presence of P2RX and TRP channels and their response to different stimuli including capsaicin, menthol, and ATP using calcium imaging. iPSC-derived human sensory neurons respond to chemotherapy drugs (ie. oxaliplatin, paclitaxel) in a dose dependent manner, showing hypersensitivity to sensory stimuli and neurotoxicity, respectively. In addition, we observed the release of Substance P and CGRP peptides in response to pain mediators. These data demonstrate the function of human iPSC-derived SNs produced at large scale, highlight their relevance for pain and neuropathy research, and showcase their utility as a tool for high-throughput drug screening for CIPN research.

Disclosures: A. Fathi: A. Employment/Salary (full or part-time); Fujifilm Cellular Dynamics Inc. Madison, WI USA. **L. Harms:** A. Employment/Salary (full or part-time); Fujifilm Cellular Dynamics Inc. Madison, WI USA. **D. Majewski:** A. Employment/Salary (full or part-time); Fujifilm Cellular Dynamics Inc. Madison, WI USA. **M. Donegan:** A. Employment/Salary (full or part-time); Fujifilm Cellular Dynamics Inc. Madison, WI USA. **K. Tomotoshi:** A. Employment/Salary (full or part-time); Fujifilm Cellular Dynamics Inc. Madison, WI USA. **S. Hilcove:** A. Employment/Salary (full or part-time); Fujifilm Cellular Dynamics Inc. Madison, WI USA. **S. Schachtele:** A. Employment/Salary (full or part-time); Fujifilm Cellular Dynamics Inc. Madison, WI USA. **J. liu:** A. Employment/Salary (full or part-time); Fujifilm Cellular Dynamics Inc. Madison, WI USA.

Poster

PSTR141. Molecular and Cellular Pain Models

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR141.04/S8

Topic: D.02. Somatosensation – Pain

Support: NIH Grant DP2NS130454

Title: Revolutionizing evoked mechanical stimulation assays through the development of the automated reproducible mechano-stimulator (ARM).

Authors: *J. BURDGE, A. JHUMKA, W. MAYISENI, I. ABDUS-SABOOR;
Columbia Univ., New York, NY

Abstract: To meet the burden posed by chronic pain, researchers have worked to identify potential analgesics using rodents and other animal models. However, there has been an increasing number of failing clinical trials. A potential cause of this translational dearth are the techniques used to measure pain in rodents, such as mechanical somatosensory assays, relying upon inconsistent and subjective human-delivered stimuli. We have revolutionized the assessment of mechanical sensitivity in rodents through the development of the Automated Reproducible Mechano-stimulator (ARM), a device delivering mechanical stimuli in a customizable and reproducible manner both in the presence of a researcher or remotely. Additional benefits include increased accessibility to scientific research and a 60% decrease in the time needed for experiments at baseline. A cohort of mice (n=10) was tested by three experimenters using pinprick stimuli delivered to the mouse paw either manually or by the ARM. Measurements using high-speed videography found an average decrease in stimulus height variation of 77.2% when using the ARM compared to manual delivery, and a corresponding 26% average decrease in the statistical variation of resulting behavioral features including max paw height, max paw velocity, and total distance traveled by the paw. Using the ARM's unique capacity to give remote mechanical stimuli we tested cohorts of male (n=10) and female (n=10) mice in the presence of either a male, female, or no researcher. Male mice showed a significant decrease in withdrawal frequency in response to cotton swab stimuli when no researcher was present compared to when either a male or female researcher was in the room. Female mice showed a significant decrease in affective pain behavior when a pinprick was applied with a female researcher present compared to no researcher. These findings indicate that the stress introduced by the presence of a researcher during somatosensory assays is enough to significantly change behavioral responses in a sex-dependent manner. Together these results indicate that the ARM is more accurate and less subjective than manual stimuli. While it makes the remote delivery of mechanical stimuli possible, it presents the potential of a promising preclinical screening tool for analgesic compounds.

Disclosures: J. Burdge: None. **A. Jhumka:** None. **W. Mayiseni:** None. **I. Abdus-Saboor:** None.

Poster

PSTR141. Molecular and Cellular Pain Models

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR141.05/S9

Topic: D.02. Somatosensation – Pain

Support: Knut and Alice Wallenberg Foundation

Title: Social avoidance of mice in pain under naturalistic conditions

Authors: *O. LE MOËNE[1], M. LARSSON[2];
[1]Linköping Univ., Linköping, Sweden; [2]Linkoping Univ., Linkoping, Sweden

Abstract: Pain includes discriminative and affective components, the latter of which can be modulated by social interactions. Reciprocally, social behavior has been suggested to be altered by pain. In particular, affective touch is thought to participate in social buffering and social contagion during aversive situations. However, pain-modulated social behavior in mice remains poorly characterized. Thus, using a novel formalin test targeting the nape of the neck, a socially relevant area, we investigated nocifensive behaviors and social interactions in group-housed mice, during the dark phase. Groups of mice (4F + 1M) were introduced to a seminatural environment (SNE) where they were left undisturbed for 6 days. On Day 5 at dusk, we first injected all mice with saline (20 µl) and observed them for 30 min (baseline). An hour after saline injection, the mice were recaptured and injected with either saline again (3 mice/group, 20 µl) or formalin (2 mice/group, 2 %, 20 µl) and observed for 30 more minutes (test). Formalin-injected (F) mice isolated themselves in the first 5 min and then displayed a steady increase in neck-scratching, inconsistent with formalin's typical 2-phase effect when administered to the paw. During the test phase, saline-injected (S) mice exhibited almost no behavioral differences compared to baseline. To the contrary, F mice emitted less social approaches and nose-off episodes during the test than at baseline. In parallel, they received less anogenital sniffing and nose-off episodes from all other mice during this phase. These differences were led by the fact that S mice emitted less of these behaviors towards F mice than towards other S mice during the test. Finally, allosniffing occurred less often between S and F mice than between S mice. In naturalistic conditions, mice in pain isolated themselves and received less pro- and anti-social behaviors from control mice in the first 30 min. To an extent, this increased social interactions between control mice. These findings do not support previous observations showing empathy in reduced behavioral mouse models, but instead indicate that in naturalistic conditions mice preferentially avoid other mice in pain.

Disclosures: O. Le Moëne: None. **M. Larsson:** None.

Poster

PSTR141. Molecular and Cellular Pain Models

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR141.06/S10

Topic: D.02. Somatosensation – Pain

Title: Acute administration of resveratrol significantly reduces acute and persistent mechanical allodynia in male and female rats; sex differences in the anti-inflammatory effect

Authors: *M. BAUTISTA CARRO[1,2], G. GALINDO-PAREDES[1,2], J. MORALES-MEDINA[2];
[1]Fisiología, Biofísica y Neurociencias, Ctr. de Investigación y de Estudios Avanzados del Inst. Politécnico Nacional, Mexico, Mexico; [2]Ctr. de Investigación en Reproducción Animal, Tlaxcala, Mexico

Abstract: Pain management is a global public health problem, especially in women, given its high prevalence and severity. Moreover, current drugs have gender differences in response to pain treatment, as well as numerous negative side effects. For these reasons, the search for new therapeutic targets for the treatment of pain is of outmost importance. Options such as resveratrol (3,5,4'-trihydroxystilbene), a phenolic compound present in many plants and fruits, as well as relatively high levels in red wine, can be very useful. Resveratrol has been shown to possess a broad spectrum of anti-inflammatory and immunomodulatory properties. In this work, we evaluated the potential antinociceptive and anti-inflammatory effects of resveratrol in two models of peripheral inflammation (acute and prolonged period) after administration of the inflammogen Complete Freund's Adjuvant (CFA) and carrageenan in male and female rats (*Rattus norvegicus*, Wistar strain). We assessed mechanical thresholds using von Frey monofilaments and determined paw thickness with a caliper. CFA or carrageenan was administered in the hindpaw to cause mechanical allodynia and paw oedema. At the peak of mechanical allodynia, resveratrol was administered orally, and mechanical thresholds and paw size were monitored until the mechanical thresholds were back to baseline values, we considered sex differences. We found that acute resveratrol treatment reverses CFA- and carrageenan-induced mechanical allodynia in male and female rats to a similar degree. Furthermore, resveratrol reduced inflammation caused by carrageenan selectively in males. We concluded that resveratrol shows promise as an effective anti-nociceptive agent in both sexes with differences in the anti-inflammatory effect.

Disclosures: M. Bautista Carro: None. **G. Galindo-Paredes:** None. **J. Morales-Medina:** None.

Poster

PSTR141. Molecular and Cellular Pain Models

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR141.07/T1

Topic: D.02. Somatosensation – Pain

Support: CINVESTAV Funding

Title: Olfactory bulbectomy induces long-lasting allodynia associated with gliosis in the amygdala in male rats

Authors: *J. MORALES-MEDINA[1], G. GALINDO-PAREDES[1], G. FLORES[2];
[1]Ctr. de Investigación y Estudios Avanzados, Tlaxcala, Mexico; [2]Benemérita Univ. Autónoma de Puebla, Puebla, Mexico

Abstract: Major depressive disorder (MDD) is an important health concern worldwide with a wide array of symptoms. Emerging evidence suggests a high comorbidity between MDD and chronic pain. Growing evidence suggest that astrocytes and microglia play a key role in both disorders. Previously, our group observed that olfactory bulbectomy (OBX) induced mechanical

and thermal cold allodynia after four weeks of surgery in male rats. At this time point, OBX induced hypertrofia in glial fibrillary acidic protein (GFAP)-positive astrocytes and hypotrofia in ionizing calcium-binding adaptor molecule 1 (Iba1)-positive microglia in the central amygdala (CeA), basolateral amygdala (BLA) and CA1 hippocampus. Remained to be investigated whether OBX induced long lasting behavioral and glial alterations important for the maintenance of chronic pain in male rats. For this reason, a battery of behavioral test was evaluated before and during 15 weeks after OBX, namely the von Frey assessment of mechanical allodynia, acetone test of thermal cold allodynia and pin prick test of mechanical hyperalgesia. At the end of the study, the brain was collected for immunohistochemistry. Our study shows that OBX induces long-lasting mechanical and thermal cold allodynia. OBX results in hypotrophy in Iba1-positive microglia in the BLA, CeA and CA1 hippocampus. OBX selectively induces hypertrophy in the CeA Moreover, while the density of Iba1-positive microglia remained similar between groups, the number of GFAP positive-astrocytes remained high in tissue from OBX rats all regions evaluated. These results suggest that glial activation results in the maintenance of chronic pain and supports the neuroinflammatory hypothesis of MDD.

Disclosures: **J. Morales-Medina:** None. **G. Galindo-Paredes:** None. **G. Flores:** None.

Poster

PSTR141. Molecular and Cellular Pain Models

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR141.08/T2

Topic: D.02. Somatosensation – Pain

Support: NIH Grant AR073187
JSPS Fellowship

Title: Contribution of Brain-derived neurotrophic factor to activity-induced muscle pain

Authors: *K. HAYASHI, J. B. LESNAK, A. N. PLUMB, A. J. JANOWSKI, A. F. SMITH, H. K. JOSLYN, K. A. SLUKA;
Univ. of Iowa, Iowa, IA

Abstract: We developed an animal model of activity-induced muscle pain. This model produces a sex-dependent pain phenotype in which females develop bilateral pain, while males develop unilateral pain. The sensory neurons play a crucial role in hyperalgesia through the activation of brain-derived neurotrophic factor (BDNF) in several animal models of pain. BDNF, a member of the neurotrophin family, is a soluble polypeptide synthesized by neuronal and non-neuronal cells and secreted into the intercellular space by autocrine, paracrine, or anterograde axonal transport. BDNF drives a pain process in spinal dorsal horn and peripheral tissues, but controversially due to the different pain model and sex. We hypothesized that pharmacological blockade of BDNF would alleviate the activity-induced muscle pain. Male and female C57BL6/J mice, aged 8 weeks were used in this study. Muscle pain was induced by 2, 20μl pH 5.0 injections, separated

by 5 days, in the gastrocnemius muscle combined with 6 minutes of electrically stimulated fatiguing muscle contractions. To assess muscle hyperalgesia, muscle withdrawal thresholds (MWT) of the gastrocnemius muscle were measured prior to and after induction of the model. Inhibitors of BDNF (ANA-12 or TrkB-Fc) were used intramuscularly or intrathecally to target the pathway. The expression of BDNF was assessed in the ipsilateral L4-L6 dorsal root ganglia (DRG) tissue and superficial spinal dorsal horn (SDH) 24hr after induction of the model using quantitative PCR and immunohistochemistry. The decrease in MWT in male, but not female, mice was reversed by intramuscular or intrathecal administration of both BDNF inhibitors 24hr after induction of the model ($p<0.05$). In parallel, at 24hr after induction of the model, there was an increase in BDNF mRNA and protein expression in DRG, when compared to controls in both male and female mice (mRNA; male: 3.1-fold, $p<0.001$; female: 3.2-fold, $p<0.001$) (protein; male: 1.5-fold, $p<0.05$; female: 1.4-fold, $p<0.05$). There was an increase in BDNF protein expression in SDH (protein; male: 1.4-fold, $p<0.05$; female: 1.3-fold, $p<0.05$), but not in mRNA, when compared to controls in both male and female mice. The current data suggests the expression of BDNF in DRG tissue after induction of the activity-induced pain were increased in both male and female mice. However, blockade of the BDNF only prevented muscle hyperalgesia in male mice; there was no effect in female mice. This data suggests there are unique mechanisms for development of activity-induced muscle pain in male and female mice.

Disclosures: K. Hayashi: None. **J.B. Lesnak:** None. **A.N. Plumb:** None. **A.J. Janowski:** None. **A.F. Smith:** None. **H.K. Joslyn:** None. **K.A. Sluka:** None.

Poster

PSTR141. Molecular and Cellular Pain Models

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR141.09/T3

Topic: D.02. Somatosensation – Pain

Support: CPRIT Grant RR210085

Title: Cancer-induced Pain in an Orthotopic Malignant Peripheral Nerve Sheath Tumor (MPNST) Mouse Model

Authors: *K. KASM[1], I. CORTEZ[2], K. IRSHAD[2], K. BAN[2], S. RASHEED[2], A. SHEPHERD[2], A. HIRBE[3], Y. PAN[2];
[1]M. D. Anderson Cancer Ctr., houston, TX; [2]Dept. of Symptom Res., M. D. Anderson Cancer Ctr., Houston, TX; [3]Washington Univ. Sch. of Med., St. Louis, MO

Abstract: Background: Cancer pain can occur at any stage of the disease development and many cancer patients experience moderate to severe pain. Despite advances in cancer treatment, pain management remains limited. Pharmacotherapy of cancer pain has relied mostly on therapeutic approaches in other pain conditions; however, the cellular and molecular mechanisms underlying cancer pain may be distinct and may require new treatment strategies. Animal models have

provided insights into the mechanisms of cancer pain, which is thought to result from complex interactions between tumor cells, the immune system, and nervous system. The use of nociceptive behavioral assays in rodent models may help to better reflect the symptoms experienced by patients. Malignant peripheral nerve sheath tumors (MPNSTs) are highly aggressive soft-tissue sarcomas arising in peripheral nerves. Patients with MPNSTs often experience pain but little is known about the mechanisms by which MPNST induces pain. Hypothesis/Goals: This study aims to characterize pain behaviors and molecular mechanism in murine MPNST models and to identify therapeutic targets. Methods: Behavioral, cell culture, and immunohistochemistry methods were performed. MPNST cancer pain model was developed by implanting the mouse MPNST cells into the sciatic nerves of either male or female mice. Primary mouse dorsal root ganglion (DRG) neuronal cell cultures were prepared for Calcium imaging experiments.Results: Mouse MPNST cell line formed tumors within the sciatic nerve following implantation. Mechanical allodynia and thermal hyperalgesia were observed in MPNST-bearing female and male mice after one week of inoculation compared to the sham groups. In addition, MPNST-conditioned medium altered calcium influx in primary cultured mouse DRG neurons, suggesting that MPNST cells secrete factors that change DRG activity.Conclusions: Using the murine MPNST allograft model, our data demonstrated that MPNST induces pain in the nerve where tumor grows. Future studies will further characterize the pain behaviors in the MPNST pain model and determine the underlying cellular and molecular mechanisms.

Disclosures: K. Kasm: None. **I. Cortez:** None. **K. Irshad:** None. **K. Ban:** None. **S. Rasheed:** None. **A. Shepherd:** None. **A. Hirbe:** None. **Y. Pan:** None.

Poster

PSTR141. Molecular and Cellular Pain Models

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR141.10/T4

Topic: D.02. Somatosensation – Pain

Support: FEDER

Title: Nociway: a new research tool to evaluate drugs in multiple pain areas in mice.

Authors: V. MAFFRE[1], L. CLAVIER[1], T. CHASSAING[1], M. FEREYROLLES[1], A. CHEFDEVILLE[1], *L. DIOP[2];
[1]ANS Biotech, RIOM, France; [2]ANS Biotech, Riom Cedex, France

Abstract: To evaluate the efficacy of exploratory compounds in different pain areas, various classical preclinical models are routinely used in mice. For early drug discovery purposes, however, the full characterization package can be long and significantly expensive. To address this issue, we have developed an innovative screening tool, named NOCIWay, a panel of behavioral pain models each validated with the most clinically relevant drugs.

Material and methods: The NOCIWay is a battery of 7 validated animal models/tests spanning a broad range of pain areas (acute and tonic pain, neuropathic pain, post-operative pain and visceral pain). The concept is an assessment of efficacy based on a group size of n=6 mice/model/test, thus providing a general pharmacological profile while reducing costs; assays/test are run in parallel, thus minimizing timelines. To validate the NOCIWay, various reference drugs classically used in clinical pain practice (morphine, oxycodone, buprenorphine, tramadol, pregabalin, acetaminophen, duloxetine, indomethacin and (-)U-50,488H) were evaluated in the 7 pain models / tests (models: CCI, post-operative; tests: electronic Von Frey, tail flick, hot plate, writhing and formalin). Behavioral and acute toxicity were also evaluated (modified Irwin grid). Results are expressed for each group as a percentage of activity for each model/test calculated from the mean value of the vehicle-treated animals from our historical database. **Results:** The effective used was reduced to n=6 by experimental groups using statistical analysis. In these experimental conditions, opioid compounds were active in all different pain models. In contrast, pregabalin and acetaminophen had more moderate efficacy. Importantly, analgesic profiles obtained with n=6 animals in the NOCIWay were in line with those generated in various and repeated fully-powered studies as well as those described in the literature.**Conclusion:** The NOCIWay provides a rapid and predictive evaluation of investigational compounds in 7 different pain models/tests, enabling their prioritization for fully-powered studies. Shortened timelines and reduced costs are possible due to small group sizes that are run largely in parallel. In summary, the NOCIWay may prove to be useful in a signal detection exercise for a broad range of potential analgesic activity.

Disclosures: **V. Maffre:** A. Employment/Salary (full or part-time); ANS Biotech. **L. Clavier:** A. Employment/Salary (full or part-time); ANS Biotech. **T. Chassaing:** A. Employment/Salary (full or part-time); ANS Biotech. **M. Fereyrolles:** A. Employment/Salary (full or part-time); ANS Biotech. **A. Chefdeville:** A. Employment/Salary (full or part-time); ANS Biotech. **L. Diop:** A. Employment/Salary (full or part-time); ANS Biotech.

Poster

PSTR141. Molecular and Cellular Pain Models

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR141.11/T5

Topic: D.02. Somatosensation – Pain

Support: NINDS grant #1R01NS115747-01A1
NIDCR grant #T90DE021984 Comprehensive Training in Inter-Disciplinary Oral Health Research
Scan Design Foundation Innovations in Pain project grant
Scan Design Innovations in Pain Summer Research Program

Title: Pleasure from pain? AS1 reverses the negative hedonic valence of aversive stimuli

Authors: *K. ESANCY, L. L. CONCEICAO, A. CURTRIGHT, T. TRAN, L. CONDON, B. LECAMP, A. DHAKA;
Biol. Structure, Univ. of Washington, Seattle, WA

Abstract: Pain is the primary reason people seek medical care, with chronic pain affecting ~ 20% of people in the USA. However, many existing analgesics are ineffective in treating chronic pain, while others (e.g., opioids) have undesirable side effects. In this study, we used a sensitized thermal place aversion assay in larval zebrafish to screen a small molecule library to identify potential novel analgesics. From this behavioral screen, we discovered a small molecule we termed Analgesic Screen 1 (AS1), which surprisingly elicited attraction to noxious stimuli. For example, when given a choice between 28.5 °C (rearing temperature) and 37.5 °C (painful heat), vehicle-treated larval zebrafish (N=58) greatly preferred their rearing temperature, spending 84.13 ± 2.15% of the total assay time in this zone (error expressed in s.e.m). By contrast, fish treated with 5 µM AS1 (N=62) only spent 31.04 ± 1.77% of the assay time in this zone, indicating preference for the normally-aversive hotter zone. AS1 was similarly able to reverse the negative hedonic valence of other aversive stimuli, eliciting attraction to both a painful chemical irritant as well as non-painful dark environments. Interestingly, AS1 did not appear to be inherently rewarding, and targeting molecular pathways canonically associated with analgesia neither replicated nor reversed the effects of AS1. A neuronal imaging assay using the neural activity marker phosphorylated-ERK revealed that distinct clusters of dopaminergic neurons, as well as forebrain regions located in the teleost equivalent of the basal ganglia, were highly upregulated in the specific context of AS1 and aversive heat. Through a combination of behavioral place preference assays and pharmacological manipulation of dopamine circuitry, we determined that AS1 acts via D1 dopamine receptor pathways to elicit attraction to a variety of noxious stimuli. Together, our results suggest that AS1 relieves an aversion-imposed "brake" on dopamine release, and that this unique mechanism may provide valuable insight into the development of new valence-targeting analgesic drugs, as well as medications for other valence-related neurological conditions, such as anxiety and post-traumatic stress disorder (PTSD).

Disclosures: K. Esancy: None. **L.L. Conceicao:** None. **A. Curtright:** None. **T. Tran:** None. **L. Condon:** None. **B. Lecamp:** None. **A. Dhaka:** None.

Poster

PSTR141. Molecular and Cellular Pain Models

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR141.12/Web Only

Topic: D.02. Somatosensation – Pain

Support: PAPIIT-UNAM Mexico Grant IN218122 to A.G-H.
PAPIIT-UNAM Mexico Grant IN202222 to M.C-L.
Fondo Sectorial de Investigación para la Educación CONACyT-Mexico Grant No. A1-S-23631 to A.G-H.
Beca de Estancias Posdoctorales por México 2022 to A. E. M. Z.

Title: Oxytocin receptor ligands (atosiban and carbetocin) inhibit early or late formalin-induced spinal nociception via agonism-biased mechanisms

Authors: *A. ESPINOSA DE LOS MONTEROS ZÚÑIGA[1], G. MARTÍNEZ LORENZANA[2], M. CONDÉS LARA[2], A. GONZALEZ-HERNANDEZ[3];
[1]Inst. de Neurobiología, UNAM, Queretaro, Mexico; [2]Univ. Nacional Autónoma De México, Queretaro, Mexico; [3]Inst. De Neurobiología, UNAM, Queretaro, Mexico

Abstract: Oxytocin receptor (OTR) at the spinal level promotes antinociception. Canonically, OTRs are coupled to the Gq proteins, but *in vitro* assays show that it also promotes the Gi pathway. Recent research suggests that spinal oxytocin blocks formalin-induced nociception via different OTR intracellular pathways. OTR-Gq inhibited early nociception (flinches), while OTR-Gi blocked late nociception (mechanical hypersensitivity). Therefore, we sought to test whether different OTR ligands could mimic this differential effect. Using the formalin test, the spinal effect of OTR-biased agonists (carbetocin or atosiban) was evaluated in male and female Wistar rats. To unravel the mechanisms implied in the effect of above biased OTR ligands, intrathecal pretreatment with L-368,899 (an OTR antagonist), U-73,122 (a phospholipase C inhibitor), L-NAME (a NOS inhibitor), or pertussis toxin (inhibitor of Gi protein function) (all given intrathecally), was used. The nocifensive behavior induced by formalin was quantified as the number of flinches (early nociception) of the injected paw during 1-h after formalin administration or paw withdrawal threshold (late nociception) for eight days after formalin administration. In male but not female rats, carbetocin only inhibits early nociception; in contrast, atosiban reduces late nociception in both sexes. The early carbetocin effect (in males) was prevented with L-368,899, U-73,122, or L-NAME, suggesting an OTR Gq-dependent pathway, whereas the atosiban-induced late antinociception (in male and female) was antagonized with L-368,899 or *pertussis* toxin, suggesting an OTR Gi-dependent pathway. These data suggest that biased OTR activation prevents early or late nociception by differential OTR pathways.

Disclosures: A. Espinosa de los Monteros Zúñiga: None. **G. Martínez Lorenzana:** None. **M. Condés Lara:** None. **A. Gonzalez-Hernandez:** None.

Poster

PSTR141. Molecular and Cellular Pain Models

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR141.13/T6

Topic: D.02. Somatosensation – Pain

Title: A high-capacity approach for identification and validation of pain targets in vitro

Authors: L. LOUHIVUORI[1], W. HENNAH[1], A. VUORENPÄÄ[1], C. STENFORS[1], L. STRID ORRHULT[2], L. MOLL[2], J. PIHL[2], *P. KARILA[2];
[1]R&D, Orion Corporation, Orion Pharma, Turku, Finland; [2]Cellectricon AB, Mölndal, Sweden

Abstract: Human genomic approaches are rapidly identifying more and more genes as significantly associated with chronic pain phenotypes. Nonetheless, for many of these genes, the physiological association to chronic pain is not evident. As changes in neuronal excitability manifest as key hall marks of chronic pain, can such targets modulate neuronal excitability? With the increasing need for new drugs for the treatment of chronic pain, a high-capacity approach to functionally identify and validate novel targets is needed.
To validate the concept, we assessed changes in neuronal excitability after lentiviral knock down (KD) of two well-established pain targets. Dorsal root ganglia neurons (DRGs) from adult rats were cultured in 384-well format for 9 and 14 days. At the day of the functional experiments, electric field stimulation (EFS) was applied to the DRGs while recording the response on an optical electrophysiology platform. The EFS response in lentiviral-treated cultures was compared with untreated controls. Cell health was evaluated by high content imaging with markers for cell nuclei, neuronal network (βIII-tubulin) and glia cells (GFAP).
Cells were transduced with lentiviral shRNA scrambled control (0.1 to 3 µl) at 1 DIV and neuronal excitability and cell health was evaluated at 9 and 14 DIV. At 9 DIV, the cultures consisted of a confluent layer of cells with a dense axonal network and EFS results were most reproducible. Moreover, there were no or small effects following treatment with lentiviral scrambled control up to a dose of 1.5 µl. These conditions were selected for lentiviral KD evaluation of pain-relevant targets.
Addition of lentiviral shRNA targeting NaV1.7 resulted in a significant decrease in excitability for three out of four oligos tested. Treatment with oligo B resulted in the strongest effect on excitability, with an SSMD score above 2. Based on the assumption that NaV1.7 is considered as a fairly strong control, an SSMD of ≥2 would be accepted as a QC criterium in a screening setting. Treatment with lentiviral shRNA targeting TrkA resulted in a significant decrease in excitability for two oligos (oligo A and B). Treatment with oligo A resulted in the strongest effect, with an SSMD above 1. Based on the assumption that TrkA is considered a moderate control, an SSMD of ≥1 would be accepted. In conclusion, we have validated an approach to assess the effect of lentiviral KD of targets on excitability in peripheral neurons and identified two suitable positive controls. Given the high capacity and relevance of the model, the platform will be suitable to identify and validate novel targets that may be of relevance for chronic pain in target identification programs.

Disclosures: **L. Louhivuori:** A. Employment/Salary (full or part-time); R&D, Orion Corporation, Orion Pharma. **W. Hennah:** A. Employment/Salary (full or part-time); R&D, Orion Corporation Orion Pharma. **A. Vuorenpää:** A. Employment/Salary (full or part-time); R&D, Orion Corporation, Orion Pharma. **C. Stenfors:** A. Employment/Salary (full or part-time); R&D, Orion Corporation, Orion Pharma. **L. Strid Orrhult:** A. Employment/Salary (full or part-time); Cellectricon AB. **L. Moll:** A. Employment/Salary (full or part-time); Cellectricon AB. **J. Pihl:** A. Employment/Salary (full or part-time); Cellectricon AB. **P. Karila:** A. Employment/Salary (full or part-time); Cellectricon AB.

Poster

PSTR141. Molecular and Cellular Pain Models

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR141.14/T7

Topic: D.02. Somatosensation – Pain

Title: A Novel Surgical Adhesion Model for Studying Endometriosis-Induced Pain and Evaluation of Celecoxib Treatment

Authors: *J. LEE, H. LEE, A. LEE, J. KWON, P. SWEENEY, S. NA, K. PARK, L. C. PARK; Naason Sci., Cheongju-si, Korea, Republic of

Abstract: Endometriosis, a condition characterized by the growth of uterine-like tissue outside the uterus, is known to cause chronic pain. It affects a significant percentage of women, with estimates ranging from 3-15% of women in their childbearing age and 25-35% of infertile women. Annually, approximately four out of 1,000 women between the ages of 15 and 64 require hospitalization due to endometriosis.While there are existing animal models for studying endometriosis, they often face limitations in accurately measuring the number of cysts. To address this challenge, our study utilized a direct surgical adhesion model. By employing this approach, we were able to observe consistently formed cysts and increased pain response in the groups with surgically implant endometriosis compared to the control groups. Furthermore, treatment with Celecoxib, a pain-relieving medication, led to an improvement in pain response assessed using the von Frey test while no significant changes in pain response were observed using the abdominal PAM test, confirming the efficacy of adhesive endometriosis in our model. To validate our novel endometriosis model, we utilized the Home Cage Analysis (HCA) system, which enables the analysis of animal behavior phenotypes. Principal Component Analysis (PCA) of the HCA system data revealed distinct differences between the control and endometriosis groups. However, no significant modifications were observed in the Celecoxib treatment group compared to the endometriosis group.In summary, our study successfully induced endometriosis and increased pain response in donor mice through the implantation of donor tissue to recipient mouse by surgical glue and modified phenotype through HCA. This newly developed model may serve as a valuable tool for further investigating the mechanisms underlying endometriosis-induced pain and exploring potential treatments for this condition

Disclosures: J. Lee: None. **H. Lee:** None. **A. Lee:** None. **J. Kwon:** None. **P. Sweeney:** None. **S. Na:** None. **K. Park:** None. **L.C. Park:** None.

Poster

PSTR141. Molecular and Cellular Pain Models

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR141.15/T8

Topic: D.02. Somatosensation – Pain

Title: Role of orexin receptors within the dentate gyrus in antinociception induced by chemical stimulation of the lateral hypothalamus in animal models of acute and persistent inflammatory pain

Authors: *A. HAGHPARAST, K. ASKARI, M. SHAREGHI BROJENI, M. RASHVAND;
Neurosci. Res. Center, Shahid Beheshti Univ. of Med. Sci., Tehran, Iran, Islamic Republic of

Abstract: Pain is a complex experience consisting of sensory, affective-motivational, and cognitive dimensions. Hence, identifying the multiple neural pathways subserving these functional aspects is a valuable task. The role of the dentate gyrus (DG) as a relay station of neocortical afferents in the hippocampal formation (HF) in the mediation of antinociceptive responses induced by lateral hypothalamus (LH)-stimulation in different animal models of pain is still a matter of controversy. Adult male Wistar rats weighing 220-250 g were unilaterally implanted with two separate cannulae into the LH and DG. Intra-DG administration of the orexin-1 receptor (OX1R) antagonist, SB334867, or the orexin-2 receptor (OX2R) antagonist TCS OX2 29 was performed just 5 min before intra-LH carbachol microinjection. Animals then underwent the tail-flick test as a model of acute pain and the formalin test using injection into the plantar surface of the hind paw as a model of persistent pain. The results showed that OX1R and OX2R antagonists dose-dependently decreased antinociceptive effects of carbachol in both tail-flick and formalin tests. In addition, the results obtained from the tail-flick test demonstrated the more prominent role of OX1R in the DG in carbachol-induced antinociception compared to that of OX2Rs in this region. According to the formalin test results, the preventive effect of SB334867 or TCS OX2 29 on carbachol-induced antinociception was approximately equal in both early and late phases of formalin nociception. Pain modulatory role of the orexinergic system through a neural pathway from the LH to DG region suggests an alternative approach to developing more efficient therapeutic agents in the clinical setting.

Disclosures: A. Haghparast: None. **K. Askari:** None. **M. Shareghi Brojeni:** None. **M. Rashvand:** None.

Poster

PSTR141. Molecular and Cellular Pain Models

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR141.16

Topic: D.02. Somatosensation – Pain

Title: Functional hiPSC-derived nociceptive sensory neurons that respond to representative nociceptor compounds and serve as models for clinical pain conditions

Authors: *K. RIEGMAN, T. WÜST, C. VAN BERKEL, S. JAIN;
Ncardia Services BV, Leiden, Netherlands

Abstract: Clinical pain conditions are a major cause for disability; an estimated 30% of individuals will experience chronic pain at some point. Nociceptive sensory neurons of the PNS are responsible for transferring nociceptive stimuli to the CNS and their aberrant activation has been linked to pain disorders in the absence of stimuli or exaggerated pain responses to non-noxious stimuli. A hallmark of these neurons is that they contain TTX-resistant sodium channel

sub-types and that they can be categorized into different ‘types’ depending on the kind of stimulus they respond to, which in turn is linked to the expression of TRP receptor sub-types. A major limitation in the field is the robust generation of hiPSC-derived nociceptive sensory neurons that contain functional channels as demonstrated by their response to representative nociceptor compounds. We generated hiPSC-derived nociceptive sensory neurons that expressed the relevant TTX-resistant sodium channel *SCN11A* and TRP receptor sub-types *TRPV1*, *TRPA1* and *TRPM8* as measured by RNAseq and ICC. To test whether these channels are functional and can be used for testing channel specific novel analgesics, we performed multi-electrode array (MEA) experiments in which we treated the neurons with TTX (targeting TTX-resistant sodium channels), Capsaicin (targeting TRPV1 channel), AITC (targeting TRPA1 channel) and Menthol (targeting TRPM8 channel). In the presence of TTX, a subset of neurons in the culture remained active and acute treatment with Capsaicin, AITC and Menthol altered activity of the neurons. Indicating that these cultures contain functional TTX resistant sodium-, TRPV1-, TRPA1- and TRPM8- channels. Diabetes is a major cause of neuropathic pain, a debilitating chronic pain disorder induced by hyperglycemia. We therefore set-out to establish a model of hyperglycemia in our functional sensory neurons. We exposed the cultures to chronic high doses of glucose and performed MEA experiments to establish the electrophysiological properties of the treated neurons. These experiments showed a difference in electrical activity as compared to control neurons. Demonstrating that a hyperglycemic state in sensory neurons can induce electrophysiological deficits and be used as a model for neuropathic pain. To conclude, we present a physiologically relevant and functional *in vitro* hiPSC-derived model of nociceptive sensory neurons, that can be used to test channel specific analgesics and demonstrated the versatility of these neurons by establishing an *in vitro* model of hyperglycemia for use in studies of neuropathic pain. This model is scalable for hit-to-lead and/or lead optimization campaigns.

Disclosures: K. Riegman: A. Employment/Salary (full or part-time); Ncardia Services BV. **T. Wüst:** A. Employment/Salary (full or part-time); Ncardia Services BV. **C. van Berkel:** A. Employment/Salary (full or part-time); Ncardia Services BV. **S. Jain:** A. Employment/Salary (full or part-time); Ncardia Services BV.

Poster

PSTR141. Molecular and Cellular Pain Models

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR141.17/T9

Topic: D.02. Somatosensation – Pain

Title: Assessing the effects of four clinically used analgesics in two mechanistic stimuli-evoked acute pain models in mice demonstrates good predictive nature of the phase 2 of formalin pain model

Authors: *H. RASHID, J. BAYOL, J. XU, S. YANG, S. BEHESHTAEIN, K. APAYART, M. BAYRAKDARIAN, Y. MAMANE;
NuChem Sci. Inc, Saint-Laurent, QC, Canada

Abstract: The objective of this study was to examine the possible predictive nature of two commonly used stimuli-evoked acute pain models in mice. Due to very high clinical attrition rates, predictive value of the preclinical pain models has been questioned in recent years. Even though several disease-related chronic pain models are currently available, the roles of stimuli-evoked acute pain models are also important due to their quick screening capabilities and their mechanistic relevance to the process of pain transmission. In this context, we have examined back-translational effects of four clinically used analgesics in two commonly used mouse acute pain models: heat-evoked tail-immersion model and chemically-evoked formalin model. In the heat-evoked tail-immersion model, the tail of the mice is immersed in a warm water bath and the time taken for reflexive withdrawal response of the tail is measured. In the chemically-evoked formalin model, diluted formalin solution is injected into mouse's paw and the time spent in nocifensive behavioral response is counted. The formalin model involves activation of peripheral nociceptors by formalin and has two distinct phases: an early direct nociceptor activation-induced phase 1 response lasting from about 0-5 minutes and a late central sensitization-induced phase 2 response lasting from about 15-60 minutes. The effects of non-steroid anti-inflammatory drug diclofenac, anti-convulsant gabapentin, sodium channel blocker mexiletine and serotonin-norepinephrine reuptake inhibitor duloxetine were examined in both models. In the tail-immersion model, only gabapentin showed slight analgesic effects at the highest dose level of 300mg/kg while diclofenac (up to 100mg/kg), duloxetine (up to 30mg/kg) and mexiletine (up to 30mg/kg) didn't show any significant analgesic effects. When these four drugs were tested in the formalin pain model at these dose levels, no significant effects were observed in the phase 1 while they produced significant analgesic effects in the phase 2 of the model, and the order of analgesia was: duloxetine > mexiletine > gabapentin > diclofenac. The effects of these drugs mainly in the second phase of formalin model is noteworthy since this phase involves a neuronal process called central sensitization, which is known to be implicated in generation of chronic pain. Hence, we suggest that the second phase of formalin model could be used as a very cost-effective and quick screening model in predicting analgesic effects of novel pain therapeutic targets in preclinical drug discovery.

Disclosures: H. Rashid: None. **J. Bayol:** None. **J. Xu:** None. **S. Yang:** None. **S. Beheshtaein:** None. **K. Apayart:** None. **M. Bayrakdarian:** None. **Y. Mamane:** None.

Poster

PSTR141. Molecular and Cellular Pain Models

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR141.18/T10

Topic: D.02. Somatosensation – Pain

Support: NIH R01 DE027223
NIH R01 DE029187

Title: Genomic profile sex-dependent differences of lingual sensory neurons in naïve and tongue-tumor bearing mice

Authors: *J. MERLO[1], T. IBRAHIM[1], P. WU[1], L.-J. WANG[2], A. TUMANOV[2], Z. LAI[2], K. WELDON[2], Y. CHEN[2], S. RUPAREL[1];
[1]Endodontics, [2]Univ. of Texas Hlth. Sci. Center, San Antonio, San Antonio, TX

Abstract: While sex-specific prevalence of orofacial pain is established, mechanisms of sex-dependent orofacial pain are widely understudied. To this end, a significant gap in knowledge exists about comprehensive regulation of tissue specific trigeminal sensory neurons in diseased state of males and females. Using RNA sequencing of FACS sorted retro-labeled sensory neurons innervating tongue tissue, we determined changes in transcriptomic profiles in male and female mice under naïve as well as tongue-tumor bearing conditions. Our data revealed the following interesting findings: 1) Tongue tissue of female mice was innervated with higher number of trigeminal neurons compared to males; 2) Naïve female neurons innervating the tongue exclusively expressed immune cell markers such as Csf1R, C1qa and others, that weren't expressed in males. This was validated by Immunohistochemistry. 3) Male neurons were more tightly regulated than female neurons upon tumor growth; 4) While very few differentially expressed genes (DEGs) overlapped between male and female post-tumor growth, several biological processes (BPs) were similar between the two sexes. However, additional distinct processes were sex-specific; 5) Post-tumor growth male DEGs contained an equal mix of transcription factors, ligands, growth factors, receptors and channels, whereas female DEGs predominantly contained channels/receptors, enzymes, cytokines and chemokines. Taken together, this is the first study to characterize the effect of sex as well as of tongue-tumors on global gene expression, biological pathways, and molecular function of tongue-innervating sensory neurons.

Disclosures: J. Merlo: None. **T. Ibrahim:** None. **P. Wu:** None. **L. Wang:** None. **A. Tumanov:** None. **Z. Lai:** None. **K. Weldon:** None. **Y. Chen:** None. **S. Ruparel:** None.

Poster

PSTR141. Molecular and Cellular Pain Models

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR141.19/U1

Topic: D.02. Somatosensation – Pain

Support: NINDS R03NS126987
PSC CUNY 64683-00-52

Title: Prrxl1 knockout -- a non-invasive model of chronic pain

Authors: E. WILLERSON[1], L. KRAMER[2], E. EICHLER[3], J. ZAR[2], K. WILAMOWSKY[3], G. CATALDO[5], ***J. C. BRUMBERG**[4];
[1]Psychology, The Grad. Center, CUNY, New York, NY; [2]Neurosci., [3]Psychology, [4]Queens College, CUNY, Flushing, NY; [5]Psychology, Queens Col., Flushing, NY

Abstract: Pain models involving invasive procedures and relatively short duration do not accurately represent the conditions of chronic pain. Prrxl1, a paired homeodomain transcription factor, is indispensable for the development of patterning in the trigeminal lemniscal pathway. In Prrxl1 knockout (KO) animals, somatotopic patterning is normal in the spinal trigeminal nucleus (SpV), yet absent along the entire trigeminal lemniscal pathway from principal sensory nucleus (PrV) to cortex. The PrV is implicated in a variety of active sensing behaviors, while the SpV is associated with transmission of noxious input. Absence of patterning is accompanied by disruption of sensory behaviors, but its involvement in nociception has not been well characterized. Prrxl1 KO animals were previously known to be hypoalgesic to the body, and we discovered that they are hyperalgesic in the orofacial region, as seen by von Frey monofilaments applied to the whisker pad and presenting behaviorally as persistent wiping of the facial region. To confirm if excessive facial grooming in Prrxl1 KO was the result of a chronic pain condition, we injected histamine, to produce itch, and capsaicin, to produce pain. When injected into the whisker pad of wildtype mice, there were two site-directed behaviors, with histamine causing scratching with the hindlimb, and capsaicin producing wiping with the forelimb - as observed in Prrxl1 KO mice. Animals were additionally assessed for hyperalgesia using the facial grimace scale, which showed that mice treated with capsaicin scored higher than animals injected with histamine or vehicle controls. We then sought to observe anatomical correlates of these behavioral conditions. Initially we assayed perineuronal nets (PNNs), lattice-like structures in the extracellular matrix, which are integral for synapse stability in the brain and spinal cord. It has been reported that PNNs appear degraded in chronic pain. PNNs in wildtype and Prrxl1 KOs were stained. We found that the PrV of Prrxl1s exhibit reduced PNNs compared with wildtype mice. We also used Iba-1 to reveal microglia whose activation has also been implicated in chronic pain. Microglia reconstructed from PrV Prrxl1 KOs took on an ‘activated’ phenotype. In sum the Prrxl1 KO mouse can be considered a chronic model of orofacial pain.

Disclosures: E. Willerson: None. **L. Kramer:** None. **E. Eichler:** None. **J. Zar:** None. **K. Wilamowsky:** None. **G. Cataldo:** None. **J.C. Brumberg:** None.

Poster

PSTR141. Molecular and Cellular Pain Models

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR141.20/U2

Topic: D.02. Somatosensation – Pain

Support: RF1NS130481

Title: Local and Systemic T Cell Dysfunction in the Tibia Fracture Model of Complex Regional Pain Syndrome

Authors: *J. WICKMAN[1], R. PANDE[2], J. DACUNZA[2], E. KASIMOGLU[2], B. B. SHENODA[3,2], S. K. AJIT[2];

[2]Pharmacol. & Physiol., [1]Drexel Univ. Col. of Med., Philadelphia, PA; [3]Intrnl. Med., Jefferson Hlth., Abington, PA

Abstract: Background: Complex regional pain syndrome (CRPS) is a debilitating chronic pain disorder with no effective treatments. Growing evidence implicates aberrant immune regulation in the induction and maintenance of CRPS pathology. Predicted targets of commonly dysregulated microRNA from CRPS patients and tibia fracture model of CRPS (TFM) mice converged on genes involved in T cell development and maintenance, including genes related to resident memory T cell (T_{rm}) function. We hypothesize that systemic T cell dysfunction in CRPS contributes to pathological T_{rm} development leading to disease development and persistence. **Methods:** Whole blood samples were obtained from CRPS patients or healthy controls. Mouse TFM was generated, and pain hypersensitivity was measured by von Frey, Hargreaves and dynamic weight bearing assays. Hind limb skin, lymph node, bone marrow, muscle, and PBMCs were collected from TFM and control mice at different time points. After enzymatic digestion and generation of single cell suspensions, T cell populations were analyzed in each tissue at different time points by flow cytometry for phenotypic and residency markers. **Results:** TFM mice demonstrate increased presence of T_{rm} populations in the skin that are phenotypically unique compared to other tissues and T_{rm} from control mice. These T_{rm} demonstrate increased $CD103^{+}$ populations. Signaling markers associated with T_{rm} development were upregulated in pathological T_{rm}. Systemic T cell dysfunction across other tissues was observed in TFM mice. Experiments evaluating the effect of pharmacological blockade of T_{rm} development signaling and depletion on TFM development, as well as Trm effector function are ongoing. **Conclusions:** Increases in pathological skin T_{rm} populations provide a novel avenue for therapeutic intervention, where blockade of signaling for T_{rm} development or systemic T cell dysfunction could prove beneficial for attenuating pain in CRPS.

Disclosures: J. Wickman: None. **R. Pande:** None. **J. Dacunza:** None. **E. Kasimoglu:** None. **B.B. Shenoda:** None. **S.K. Ajit:** None.

Poster

PSTR142. Inflammatory Pain

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR142.01/U3

Topic: D.02. Somatosensation – Pain

Support: NIH Grant 4R00NS11410703

Title: Investigating the Central Mechanism Controlling Pain Sensitivity During Acute Inflammation

Authors: *J. NGUYEN, M. SLATER, J. A. OSTERHOUT;
Neurobio., Univ. of Utah, Salt Lake City, UT

Abstract: Infection is a global health concern, with accompanying sickness symptoms that aid to combat the invading pathogens. While the neurobiology of certain sickness symptoms during infection has been explored, the neural mechanisms underlying increased pain perception and pain-related behavioral responses in the context of acute inflammation remain poorly understood. Here we aim to uncover the central neurons and neural circuits involved in inflammation-induced hyperalgesia and motivated pain behaviors. Using cutting-edge approaches for cell type identification and behavioral analysis we have identified candidate neuronal populations that are critical for pain sensitivity and associated behaviors following administration of pro-inflammatory lipopolysaccharides. Functional manipulation of these populations reveal their critical role in the generation of increased pain sensitivity and pain-related behaviors during infection. Together, these results are vital for understanding central neural circuits required for sickness behaviors as well as identifying potential targets for novel pain therapeutics and mitigating the negative consequences associated with chronic pain.

Disclosures: J. Nguyen: None. **M. Slater:** None. **J.A. Osterhout:** None.

Poster

PSTR142. Inflammatory Pain

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR142.02/U4

Topic: D.02. Somatosensation – Pain

Support: Tri Service Nursing Research Program USUHS Award NA20-A02GR
CDMRP grant W81XWH-21-2-001

Title: Reversal of mechanical hypersensitivity in mice by (2R,6R)-hydroxynorketamine (HNK) requires activation of group 2 metabotropic glutamate receptors

Authors: *K. R. CASTELL[1,2], M. A. CAMPANILE[1,2], J. O. PAMPALONE[1,2], C. A. BROWNE[2,1], I. LUCKI[2,3];
[1]Henry M. Jackson Fndn. for the Advancement of Military, Bethesda, MD; [2]Pharmacol. and Mol. Therapeut., [3]Psychiatry, Uniformed Services Univ., Bethesda, MD

Abstract: 2R,6R)-hydroxynorketamine (HNK) exerts analgesic action in several rodent pain models. This study evaluated the contribution of Group 2 metabotropic glutamate receptors (mGlu2 and mGlu3) to alter HNK-mediated reversal of mechanical hypersensitivity in a murine inflammatory pain model with either a selective mGlu2/3R agonist LY379268 (LY68), or an mGlu2R negative allosteric modulator, VU6001966 (VU66). Ninety-six adult male C57BL/6J mice received a 20µl intra-plantar injection of 2.5% λ carrageenan into the left hind paw to induce mechanical hypersensitivity. LY379268 (10 mg/kg; experiment 1), VU6001966 (10 mg/kg; experiment 2), or vehicle were administered intraperitoneally 90 minutes post-carrageenan and 30 minutes prior to HNK (30 mg/kg). Hypersensitivity was measured using Von Frey filaments 4 and 24 hours after HNK. Scores were reported as percent return to baseline. At

each time point, two-way ANOVA determined Pretreatment x Treatment interactions, with Šídák's multiple comparisons conducted where appropriate. At 4 hours, carrageenan induced mechanical hypersensitivity, with scores at 15% of baseline values in saline/vehicle treated mice. HNK reversed mechanical hypersensitivity in experiment 1 ($F_{1,40} = 14.13$, $P=0.0005$) and 2 ($F_{1,40} = 4.533$, $P=0.0394$), return to baseline values, 26% and 32% respectively. No significant pretreatment/treatment interactions were evident at this time point. Mechanical hypersensitivity persisted at 24 hours (14% of experiment 1 baselines and 24% of experiment 2 baselines). HNK induced robust analgesia in Experiment 1 ($F_{1,41} = 16.13$, $P=0.0002$) and 2 ($F_{1,41} = 9.744$, $P=0.0033$). LY68 alone also reversed mechanical hypersensitivity ($F_{1,41} = 5.2088$, $P=0.0277$). Combined LY68/HNK treatment enhanced return to baseline values from 48 to 55%. Conversely, VU66 blocked HNK's effects (Pretreatment x Treatment interaction; $F_{1,41} = 4.68$, $P=0.0362$), with mean return to baseline values of 45% in Vehicle/HNK animals compared to VU66/HNK mice at 25%. These data support the hypothesis that mGluR2s may contribute to HNK's analgesic action. Acknowledgements We are grateful to Dr. Craig J. Thomas of the National Center for Advancing Translational Sciences for supplying (2R,6R)-hydroxynorketamine and the USU Preclinical Behavioral and Modeling core for the use of the center's equipment Disclaimer: The content and conclusions do not necessarily represent the official position or policy of the Uniformed Services University of the Health Sciences, the Department of Defense, or the U.S. Government. The authors declare no conflicts of interest

Disclosures: **K.R. Castell:** None. **M.A. Campanile:** None. **J.O. Pampalone:** None. **C.A. Browne:** None. **I. Lucki:** None.

Poster

PSTR142. Inflammatory Pain

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR142.03/U5

Topic: D.02. Somatosensation – Pain

Support: CIC UMSNH 16060
CIC UMSNH 16124
CIC UMSNH 16125
ICTI PICIR-075

Title: Analgesic metformin-melatonin interaction in the rat formalin test

Authors: J. MARTINEZ-GUILLEN[1], D. GODINEZ-HERNANDEZ[2], C. J. GUTIERREZ-GARCIA[3], M. Y. GAUTHEREAU-TORRES[1], *L. F. ORTEGA-VARELA[4];
[1]1.Facultad de Ciencias Médicas y Biológicas "Dr. Ignacio Chávez", [2]IIQB, Univ. Michoacana de San Nicolas de Hidalgo, Morelia, Mexico; [3]Dept. de Ingeniería química y Bioquímica, Tecnológico Nacional de México/Instituto Tecnológico de Morelia, Morelia, Mexico; [4]Salud Publica. Univ. Michoacana Sn Nicolas De H ., Morelia, Mexico

Abstract: Pain is a growing public health problem worldwide. The combination of drugs is highly used in analgesic therapy, in order to improve the analgesic effects and decrease its adverse effects. Drugs with pleiotropic effects such as metformin (antidiabetic) and melatonin (sleep regulator), have potential to act as an efficient combination. This study was achieved to assess the interaction between metformin and melatonin orally administered in the rat model of formalin test. Female Wistar rats (220-350 g) were injected into the dorsal surface of the right hind paw with 50 microliters of 1% formalin. This substance induced a flinching pain-related behavior, the reduction of such conduct is considered as antinociception. The percent of antinociceptive effect was determined by the oral administration of metformin (30-1000 mg/kg), melatonin (10-150 mg/kg), and their combination. To stablish the nature of the interaction, isobolographic analysis was used in a fixed-dose radio (0.5:0.5), on the basis of their ED50 values: metformin (947.46 ± 242.60 mg/kg) and melatonin (126.86 ± 37.98 mg/kg). The metformin-melatonin combination significantly reduced the number of flinches in the second phase of the formalin test. The theoretical effective dose 50 for the combination (ED50 T) was 537.15 ± 122.76 mg/kg. Experimentally, the effective dose 50 (ED50 E) was significantly lower (360.83 ± 23.36 mg/kg), indicating synergistic effects for the combination. Results show that the oral coadministration of metformin and melatonin could provide a therapeutic alternative for inflammatory pain.

Disclosures: J. Martinez-Guillen: None. **D. Godinez-Hernandez:** None. **C.J. Gutierrez-Garcia:** None. **M.Y. Gauthereau-Torres:** None. **L.F. Ortega-Varela:** None.

Poster

PSTR142. Inflammatory Pain

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR142.04/U6

Topic: D.02. Somatosensation – Pain

Support: Prof. KH René Koczorek Stiftung, Neuried, Germany.

Title: Steroid receptor-mediated alterations in the genomic regulation of pain signaling molecules of primary afferent neurons.

Authors: *S. MOUSA[1], S. MOHAMMED[1], M. SHAKIBAEI[2], A. BEYER[3], S. TRESKATSCH[1], M. SCHÄFER[1];
[1]Dep. of Anesthesiology, Charite Univ., Berlin, Germany; [2]2Depart. of Anat., Ludwig-Maximilians-University Munich, Munich, Germany; [3]Dept. of Anaesthesiology, Ludwig-Maximilians-University Munich, Munich, Germany

Abstract: Accumulating morphological and functional data suggested mineralocorticoid receptors are a promising target for pain relief. According to the classical genomic response of steroid receptors, steroids activate their receptors located in the cytoplasm to trigger the transcription of their target genes including presumably pain signaling molecules. A most recent

study suggested that mineralocorticoid and glucocorticoid receptor ligands can modulate pain perception by a distinct genomic response. Therefore, we investigated changes in the genomic expression of specific pain signaling molecules within cultured PC12 and N18TG2 cell lines in vitro as well as in dorsal root ganglion peripheral sensory neurons in vivo. The expression of putative pain signaling molecules such as TRPV1, Nav1.8, CGRP, and trkA were evaluated under the influence of mineralocorticoid and glucocorticoid receptor agonists and antagonists by quantitative Real-Time Polymerase Chain Reaction (RT-PCR) and double immunofluorescence confocal microscopy. Here, our RT-PCR as well as immunofluorescence confocal microscopy analysis demonstrated that distinct genes of pain signaling molecules were differentially up- or downregulated under the influence of mineralocorticoid and glucocorticoid receptor agonists and antagonists in vitro. Similarly, the in vivo experiments showed a similar pattern of the genomic expression profile of pain signaling molecules in primary afferent neurons under the influence of mineralocorticoid and glucocorticoid receptor ligands. Taken together, these preliminary findings may increase our understanding of receptor-mediated regulation of gene transcription with specific regard to pain signaling molecules within peripheral sensory neurons and this may be useful for improving pain management using steroids. Supported by the Prof. KH René Koczorek Stiftung, Neuried, Germany.

Disclosures: **S. Mousa:** None. **S. Mohammed:** None. **M. Shakibaei:** None. **A. Beyer:** None. **S. Treskatsch:** None. **M. Schäfer:** None.

Poster

PSTR142. Inflammatory Pain

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR142.05/U7

Topic: D.02. Somatosensation – Pain

Support: BBSRC PhD Studentship BB/M011194/1
MRC Grant MR/W002426/1
BBSRC/GSK iCASE PhD Studentship BB/V509528/1

Title: A non-neuronal role for the proton-sensing GPCR, GPR65 in inflammatory joint pain.

Authors: *L. A. PATTISON[1], R. H. RICKMAN[1], H. HILTON[1], S. WIJESINGHE[2], G. LADDS[1], L. YANG[3], S. JONES[2], E. S. SMITH[1];
[1]Dept. of Pharmacol., Univ. of Cambridge, Cambridge, United Kingdom; [2]Inst. of Inflammation and Ageing, Univ. of Birmingham, Birmingham, United Kingdom; [3]Dept. of Intrnl. Medicine, Brody Sch. of Med., East Carolina Univ., Greenville, NC

Abstract: Therapeutic options for treating inflammatory pain are limited and plagued by side-effects. Localized acidosis is characteristic of inflammation and many receptors expressed by pain-sensing neurons (nociceptors) are sensitive to extracellular protons, which makes them potential targets for treating inflammatory pain. While proton-sensitive ion channels have more

established roles in nociception, less is known about the roles of proton-sensing G protein-coupled receptors (PS-GPCRs). In several inflammatory pain models increased expression of PS-GPCRs has been reported. In particular, GPR65 (also known as TDAG8) expression often increases following inflammatory insult, and thus we sought to further explore the role of GPR65 in inflammatory pain. First, in a uniform cellular background the signaling responses elicited following GPR65 activation by protons and two other reported agonists, BTB09089 and psychosine, were compared. BTB09089 best recapitulated acid-induced signaling, highlighting its use as a more-selective tool to probe GPR65 function. Injection of BTB09089 into the mouse knee joint caused inflammation (BTB-injected knee width: 3.60 ± 0.20 mm versus DMSO-injected knee: 3.04 ± 0.14 mm) and pain-related behaviors in mice of both sexes. Additionally, knee-innervating sensory neurons supplying the BTB09089 injected knee were hyperexcitable (Ipsilateral side rheobase: 320.9 ± 44.0 pA versus Contralateral side: 562.7 ± 47.6 pA). However, this could not be re-created by directly stimulating sensory neurons isolated from naïve mice with BTB09089, thus suggesting the involvement of another cell type. Fibroblast-like synoviocytes (FLS) resident in the joint also express GPR65. Stimulation of FLS with BTB09089 increased cAMP accumulation and induced secretion of inflammatory mediators capable of sensitizing naïve sensory neurons. The specificity of these results was confirmed through studies with $GPR65^{-/-}$ mice. Similar effects of BTB09089 on FLS isolated from arthritic human patients were also observed. We thus postulate that GPR65 is a key mediator of cell-cell interactions responsible for the manifestation of inflammatory pain in both humans and mice, and thus of interest in the development of more targeted therapeutics for treating inflammatory diseases.

Disclosures: L.A. Pattison: None. **R.H. Rickman:** None. **H. Hilton:** None. **S. Wijesinghe:** None. **G. Ladds:** None. **L. Yang:** None. **S. Jones:** None. **E.S. Smith:** None.

Poster

PSTR142. Inflammatory Pain

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR142.06/U8

Topic: D.02. Somatosensation – Pain

Support: MOST 111-2320-B-A49-011-MY3
MOST 109-2320-B-010-027-MY3
MOST 111-2740-B-001-002-

Title: Middle-aged female mice with high pro-inflammatory cytokine level develop severe symptoms in the early stage of Rheumatoid arthritis

Authors: *C.-W. LO[1,3], W.-H. SUN[3], C.-C. CHEN[1,2];
[1]Taiwan Mouse Clinic, Biomed. Translational Res. Ctr., [2]Inst. of Biomed. Sci., Academia Sinica, Taipei, Taiwan; [3]Dept. of Life Sci. and Inst. of Genome Sci., Natl. Yang Ming Chiao Tung Univ., Taipei, Taiwan

Abstract: Rheumatoid arthritis (RA) is a chronic autoimmune disease with joint synovium of inflammation, which induces joint deformation, cartilage loss and pain. Although anti-inflammatory drugs have developed and applied on RA patients, there are still many RA patients who do not respond to current treatments. Pain relief is one of the top priorities in RA patients. Clinical studies found that middle-aged male RA patients have better responses after drug treatments. The differences between male and female could be attributed to the differences in the immune system. Some studies found that women have higher activated T cells number, and higher Type I interferon activity than men. However, it remains unclear whether the sexual dimorphism of RA symptoms only occurs in middle-aged RA patients and whether this sexual differences are related to the immune system. We intra-articularly injected Complete Freund's Adjuvant (CFA) into 2-, 10-, 18-month-old male and female mice to induce RA development, followed by arthritis severity assessment, pain behavioral tests, H&E and cytokine analysis. We found that the 10-month-old female RA mice developed more severe pain than 10-months-old male RA mice in early stage (1-4w). These results were associated with higher synovial inflammation in 10-month-old female than male mice. When compared with 10-month-old male mice, female mice had a higher number of non-granular immune cells and high expression of proinflammatory cytokines. Thus, more severe pain and inflammation observed in the early stage of RA in middle-aged female mice could be associated with the sexual difference in immune system. Further mechanism causes such difference will be studied later.

Disclosures: C. Lo: A. Employment/Salary (full or part-time); Academia Sinica/full. **W. Sun:** A. Employment/Salary (full or part-time); National Yang Ming Chiao Tung University/full. **C. Chen:** A. Employment/Salary (full or part-time); Academia Sinica/full.

Poster

PSTR142. Inflammatory Pain

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR142.07/U9

Topic: D.02. Somatosensation – Pain

Support: Grant Agency of the Czech Republic 21-02371S

Title: Cb1r mediated inhibition of nociceptive trasmission in the spinal cord is dependent on sgip1 protein

Authors: *M. PONTEARSO[1], A. BHATTACHARYA[1], J. SLEPICKA[1], D. MUZIK[2], D. DRESSLEROVA[2], J. PALECEK[1], D. SPICAROVA[1];
[1]Inst. of Physiol. - CAS, Prague, Czech Republic; [2]Fac. of Sci., Charles Univ., Prague, Czech Republic

Abstract: CB1R mediated inhibition of nociceptive transmission in the spinal cord is dependent on SGIP-1 protein
Monica Pontearso, Anirban Bhattacharya, Jakub Slepicka, David Muzik, Denisa Dresslerova,

Jiri Palecek, Diana Spicarova
Laboratory of Pain Research, Institute of Physiology Czech Academy of Sciences, , Prague, Czech RepublicCharles University, Physiology Department, Prague
Chronic pain is a debilitating condition that affects a significant part of the worldwide population and is one of the most common reasons to seek medical care. Cannabinoid receptor 1 (CB1R) is one of the pivotal receptors in modulating nociceptive synaptic transmission at the spinal cord level in normal and pathological states. In synapses, CB1R signalling is regulated by the protein Src homology 3-domain growth factor receptor-bound 2-like (SGIP1), limiting receptor internalization while enhancing association with β-arrestin. We investigated the role of SGIP1 in nociceptive synaptic transmission at the first synapse of the pain pathway in the spinal cord dorsal horn under control and inflammatory conditions. Patch-clamp recordings of miniature excitatory postsynaptic currents (mEPSCs) from superficial dorsal horn neurons in mouse spinal cord slices and calcium imaging experiments in dorsal root ganglia neurons were performed in SGIP1 knock-out (KO) and wild-type (WT) mice. Electronic Von Frey and Hot Plate behavioral tests were used to assess mechanical and thermal sensitivity. Peripheral inflammation was induced by the intraplantar application of carrageenan into the hindpaw. The KO mice showed significantly lower sensitivity to the heat stimulus than the wild-type mice. The inhibition of mEPSC frequency induced by CB1R agonist WIN 55,212-2 application was significantly higher in KO (42.3 %) than in WT (85.9%) after induction of peripheral inflammation. In naïve animals, the inhibition induced by WIN 55,212-2 application was similar in the SGIP1 KO and WT. Increased effectiveness of WIN 55,212-2 application was also demonstrated on the excitability of DRG neurons in KO mice. In conclusion, SGIP1 KO mice showed lower sensitivity to thermal stimuli than SGIP1 WT mice under control conditions. Under inflammatory conditions, the CB1R agonist application was more effective in inhibiting nociceptive synaptic transmission in SGIP1 KO than in WT mice. Our results suggest that SGIP1 interaction with CB1R on primary afferent terminals in the spinal cord dorsal horn could play an important role in nociceptive transmission, especially under inflammatory conditions.
Supported by the Grant Agency of the Czech Republic 21-02371S.

Disclosures: **M. Pontearso:** None. **A. Bhattacharya:** None. **J. Slepicka:** None. **D. Muzik:** None. **D. Dresslerova:** None. **J. Palecek:** None. **D. Spicarova:** None.

Poster

PSTR142. Inflammatory Pain

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR142.08/U10

Topic: D.02. Somatosensation – Pain

Title: The CaCCs channels Bestrophin-1 and Anoctamin-1 are involved in the mechanical allodynia induced by REM sleep deprivation in rats

Authors: *C. MARTÍNEZ MAGAÑA, P. MUÑOZ CASTILLO, J. MURBARTIAN; Farmacobiología, Ctr. de Investigación y Estudios Avanzados, Ciudad de México, Mexico

Abstract: Bestrophin-1 and anoctamin-1 (also known as TMEM16A) belong to calcium-activated chloride channels (CaCCs) family. Recently, several studies showed that these channels are expressed in DRG (dorsal root ganglia) neurons and are involved in nociception induced by inflammatory factors such as bradykinin and also in neuropathic pain. However, their role in pain hypersensitivity induced by REM sleep deprivation (REMSD) has not been studied. The aim of this study was to determine if bestrophin-1 and anoctamin-1 are involved in the maintenance of tactile allodynia induced by REM sleep deprivation. We use a multiple platform method for inducing REMSD and evoke mechanical allodynia in rats. The REMSD for 48 h induced tactile allodynia, & a transient increase in plasmatic corticosterone concentration at the beginning of the protocol (12 h) in female and male rats. Compared to vehicle-treated rats, intrathecal injection of $CaCC_{inh\text{-}A01}$, a blocker of bestrophin-1, & $T16A_{inh\text{-}A01}$, a specific anoctamin-1 blocker, temporarily relieved REMSD-induced tactile allodynia in both sexes. However, $T16A_{inh\text{-}A01}$ had a higher antiallodynic effect in male than female rats. The western blot analysis reveals that REMSD up-regulated bestrophin-1 protein expression in dorsal root ganglia (L4 & L5 pool) but no changed its expression in dorsal spinal cord (DSC) in male & female rats. In marked contrast, REMSD decreased anoctamin-1 protein expression in DSC only in female rats, but no changes were observed in DRG. These data suggest that bestrophin-1 and anoctamin-1 have a pronociceptive role in the maintenance of tactile allodynia induced by REM sleep deprivation in rats, but the changes in ANO1 protein expression in spinal cord are different between sexes, which could explain the greater sensitivity of males to ANO1 blocker.

Disclosures: C. Martínez Magaña: None. **P. Muñoz Castillo:** None. **J. Murbartian:** None.

Poster

PSTR142. Inflammatory Pain

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR142.09/V1

Topic: D.02. Somatosensation – Pain

Support: NIH Grant R01- NS121259
NIH Grant R01- DK103808
NIH Grant T32-GM092715

Title: Lateral Hypothalamic Area Neurons Expressing Neurotensin Support Weight Loss and Alleviate Pain

Authors: *R. KHAN, G. BEENHWA, K. INYANG, R. BUGESCU, E. TANUSHI, H. BEMIS, G. LAUMET, G. LEINNINGER;
Michigan State Univ., East lansing, MI

Abstract: Chronic pain and obesity frequently occur together. An ideal therapy would alleviate pain without weight gain, and most optimally, could promote weight loss. The neuropeptide neurotensin (Nts) is implicated in reducing weight and pain, but the endogenous mechanisms

underlying this physiology were unknown. We previously showed that activating lateral hypothalamic area neurons expressing Nts (LHA^{Nts} neurons) suppresses feeding and promotes weight loss. Here we hypothesized that activating LHA^{Nts} neurons can also alleviate pain. To test this, we injected Nts^{Cre} mice in the LHA with AAVs to cre-dependently express either mCherry (Control) or excitatory Designer Receptors Exclusively Activated by Designer Drugs (DREADDs) in LHA^{Nts} neurons, permitting their activation after treatment with the DREADD ligand clozapine N-oxide (CNO, 0.3 mg/kg, i.p.). Activating LHA^{Nts} neurons had no effect on thermal pain responses in naïve mice. By contrast, spared nerve injury-induced pain hypersensitivity was completely reversed by CNO-mediated activation of LHA^{Nts} neurons compared to VEH control, both acutely and after chronic injury. In mice treated with complete Freund's adjuvant (which induces inflammatory pain), activating LHA^{Nts} neurons also relieved pain hypersensitivity. However, pretreatment with the brain permeable Nts receptor pan-antagonist SR142948 (1mg/kg, i.p, 30 min before VEH/CNO) blocked CNO-mediated analgesia, indicating that LHA^{Nts} neurons alleviate chronic pain in an Nts-dependent manner. Excitingly, activating LHA^{Nts} neurons in diet-induced obese mice alleviated their baseline and CFA-induced pain. Taken together these data suggest that augmenting signaling via LHA^{Nts} neurons may be a common actionable target for both pain and obesity.

Disclosures: R. Khan: None. **G. Beenhwa:** None. **K. Inyang:** None. **R. Bugescu:** None. **E. Tanushi:** None. **H. Bemis:** None. **G. Laumet:** None. **G. Leinninger:** None.

Poster

PSTR142. Inflammatory Pain

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR142.10/V2

Topic: D.02. Somatosensation – Pain

Support: NIH Grant ZIA DE-000664

Title: Transcriptome Analysis of Painful Rheumatoid Arthritis

Authors: B. HALL[1], C. JUDKINS[1], M. CASSIDY[1], E. MACDONALD[1], A. TERSE[1], S. YUN[1], ***A. B. KULKARNI**[2];
[1]NIH, Natl. Inst. of Hlth., Bethesda, MD; [2]NIH, Natl. Inst. of Dent. and Craniofacial Res., Bethesda, MD

Abstract: Autoimmune diseases such as rheumatoid arthritis (RA) can cause states of chronic inflammation with accompanying tissue destruction and pain. To better understand the mechanisms and consequences of ongoing inflammatory-induced pain signaling, dorsal root ganglia (DRG) were acquired from individuals with RA for transcriptomic and neuroanatomical study. We conducted RNA sequencing from the L5 DRGs because it contains the soma of the sensory neurons that innervate the foot, where 80-90% of RA patients report foot problems. DRGs from 5 RA patients were compared with 9 non-arthritic controls. RNAseq of L5 DRGs

identified 128 differentially expressed genes (DEGs) that were dysregulated in the RA subjects as compared to the non-pain controls, of which 52 were protein-coding genes. The DRG resides outside the blood brain barrier and, as such, our initial transcriptome analysis detected signs of an autoimmune disorder including the upregulated expression of immunoglobulins and other immunologically related genes within the RA individuals. Additionally, we saw the upregulation in genes implicated in neurogenesis that could promote pain hypersensitivity. Overall, our DRG analysis suggests that there may be upregulated inflammatory and pain signaling pathways that may contribute to chronic pain.

Disclosures: B. Hall: None. **C. Judkins:** None. **M. Cassidy:** None. **E. Macdonald:** None. **A. Terse:** None. **S. Yun:** None. **A.B. Kulkarni:** None.

Poster

PSTR142. Inflammatory Pain

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR142.11/V3

Topic: D.02. Somatosensation – Pain

Support: NIH R01 NIDA 044999-01A1S1
NIH R01 NIDA 044999-01A1
Grant 121035 Texas Tech University

Title: Assessment of dose- and sex-differences in antinociception mediated by cannabidiol and amitriptyline with exploration of a shared serotonin 1A receptor mechanism of combined antinociception

Authors: *R. C. BARNES[1], S. BANJARA[1], I. CASTRO-PIEDRAS[1], D. J. MORGAN[2], J. GUINDON[1];
[1]Pharmacol. and Neurosci., Texas Tech. Univ. Hlth. Sci. Ctr., Lubbock, TX; [2]Biomed. Sci., MARSHALL UNIVERSITY, HUNTINGTON, WV

Abstract: Objectives: As the prevalence of chronic pain continues to rise, exploration of new pain therapies grows in importance. Cannabidiol (CBD) is found in cannabis and has recently received both renewed interest for use as an analgesic. Amitriptyline (AT) is a tricyclic antidepressant that has already received approval for use in chronic pain conditions, though much its research excluded female subjects. We sought to elucidate possible sex differences in CBD and AT analgesia using C57BL/6j mice in the formalin model of inflammatory pain. Additionally, we explored the benefit of combining these drugs through a shared mechanism involving 5-HT_{1A} serotonergic receptors. **Methods:** This study was performed in adult male and female wild-type C57BL/6j mice. Mice were pretreated via intraperitoneal injection with either vehicle, CBD (0.3, 1, 2.5, 10, 30, and 100 mg/kg), or AT (0.1, 0.3, 1, 3, 10, and 30 mg/kg) and then allowed to adapt for 20 minutes. Mice were then injected with 10 µL of 2.5% formalin subcutaneously into their hind paw and pain behavior was scored for one hour. Results were used

to find the median effective dose (ED_{50}). Further mice were pretreated with ED_{50} dose of either CBD, AT, or both. A final group received pretreatment with selective 5-HT_{1A} antagonist WAY100635 20 minutes prior to injection with either a vehicle or both CBD and AT ED_{50} doses. **Results:** CBD and AT both proved antinociceptive in both the acute and inflammatory phases of the formalin test. The dose required to achieve significance was lower in the inflammatory phase and in males compared to females for both drugs (for the inflammatory phase, significance was achieved for CBD at 10 mg/kg in females and at 2.5 mg/kg in males and for AT at 1 mg/kg in females and at 0.3 mg/kg in males). Both CBD and AT were found to have significant sex differences. Both male and female mice saw a significant additive effect in the acute phase of the formalin test with the combination of ED_{50} doses. Only male mice demonstrated this effect in the inflammatory phase. Pretreatment with WAY-100635 completely reversed the acute phase antinociception, and partially reversed the inflammatory phase antinociception, mediated by the combination of CBD and AT ED_{50} doses. **Conclusions:** This study demonstrates significant sex differences in AT and CBD analgesia, highlighting the need for further research into this sex specific effect and potential therapeutic relevance. Additionally, the discovery of benefit in combining these drugs supports their combination in a multimodal approach to pain. Further research is ongoing to analyze changes in gene expressions caused by treatment with these compounds in the formalin model of inflammatory pain.

Disclosures: R.C. Barnes: None. **S. Banjara:** None. **I. Castro-Piedras:** None. **D.J. Morgan:** None. **J. Guindon:** None.

Poster

PSTR142. Inflammatory Pain

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR142.12/V4

Topic: D.02. Somatosensation – Pain

Support: VA Merit Review BX005899
VA Merit Review BX006244

Title: Microglial and Macrophage Deletion of the RNA Regulator Tristetraprolin Enhances Postoperative Incisional Pain through Activation of Neuroinflammatory Responses

Authors: *A. GUHA[1,2,4], R. SORGE[3], Y. SI[1,2,4], R. SMITH[1,2], P. BLACKSHEAR[5], P. KING[1,2,4];
[1]Neurol., [2]Ctr. for Neurodegeneration and Exptl. Therapeut., [3]Psychology, Univ. of Alabama, Birmingham, Birmingham, AL; [4]Neurol., Birmingham VA Med. Ctr., Birmingham, AL; [5]Signal Transduction Lab., Natl. Inst. of Envrn. Hlth. Sci., Research Triangle Park, NC

Abstract: Pro-inflammatory activation of microglia and infiltrating macrophages in the central nervous system (CNS) is a major driver of disease progression in many neurological disorders, ranging from acute traumatic injury to chronic neurodegenerative diseases. A hallmark feature of

this activation is production of pro-inflammatory factors such as IL-6, TNF-α, IL-1β, and iNOS. Posttranscriptional regulation via AU/U-rich elements (ARE) in the mRNAs encoding these factors is a major control point for their expression in microglia and macrophages. We recently reported that the ARE-RNA binding protein, Human antigen R (HuR), positively regulates production of these pro-inflammatory mediators in activated microglia, and that its inhibition attenuates neuroinflammation and mitigates neuropathic pain (NP) after peripheral nerve injury. Tristetraprolin (TTP) is another ARE-RNA binding protein that binds to the same target mRNAs, but functions as a negative regulator by promoting mRNA degradation. Since inflammation is a major driver of neuropathic pain after peripheral injury, we hypothesized that deletion of TTP in microglia/macrophages may enhance inflammation and pain in a surgical incision model. Using a macrophage/microglial TTP knockout (KO) mouse model, we found that TTP deletion exacerbates mechanical allodynia, a hallmark of NP, after surgical incision. We assessed mRNA expression of inflammatory mediators in skin near the surgical incision site, lumbar dorsal root ganglia (DRG), and lumbar spinal cord. At the incision site, there was a robust induction of pro-inflammatory mediator mRNAs, including *IL-1b*, *IL-6*, *TNF-α, iNOS*, *COX2*, *CCL2*, *CXCL1* and *CXCL2*, that was significantly enhanced in TTP KO mice. In DRG ipsilateral to the incision, as well as lumbar spinal cord, there was increased mRNA expression of select pro-inflammatory mediators in TTP KO mice. IL-10 mRNA was not altered by TTP KO at any level. In summary, TTP plays a critical role in tamping down peripheral and CNS pro-inflammatory responses that drive allodynic pain after surgical incision. Enhancing TTP function may represent a new direction in the treatment of neuropathic pain and other disorders driven by neuroinflammation.

Disclosures: A. Guha: None. **R. Sorge:** None. **Y. Si:** None. **R. Smith:** None. **P. Blackshear:** None. **P. King:** None.

Poster

PSTR142. Inflammatory Pain

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR142.13/V5

Topic: D.02. Somatosensation – Pain

Support: 2020R1C1C101024513
2022M3E5E801739512
KIST Grant 2E32231

Title: Piezo1 transduces inflammatory pain signals in nociceptors

Authors: *T. HA[1,2], P. LEE[1], G.-S. HONG[1];
[1]Korea Inst. of Sci. and Technol., Seoul, 02792 Republic of Korea, Korea, Republic of; [2]Korea Univ., Seoul, Korea, Republic of

Abstract: Mechanosensation begins with the sensing of pressure by mechanically activated (MA) channels in the nerve endings of dorsal root ganglion (DRG) neurons. Piezo1, a

fastinactivating MA channel, has surfaced to be involved in pruriception. However, the pressure-dependent activation mechanism and its physiological role in mechanical pain remain unidentified. Here, we report that Piezo1 is expressed in a small DRG subpopulation, which is largely positive for TRPV1 rather than MRGPRD, which is known for nociceptors. To investigate the molecular function of Piezo1 in DRG neurons, we reclassified DRG neurons based on the MA current type. The silencing of the Piezo1 gene resulted in two subgroups—intermediately adapting (IA) and intermediately slowly adapting (ISA) responders of DRG neurons. Silencing Piezo1 in mice via specific lumbar DRG-targeted ganglionic injection of shRNA virus reduced tactile pain hypersensitivity in formalin- and carrageenan-dependent inflammation. Piezo1 mediates mechanical pain by acting as a nociceptive MA channel

Disclosures: **T. Ha:** None. **P. Lee:** None. **G. Hong:** None.

Poster

PSTR142. Inflammatory Pain

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR142.14/V6

Topic: D.02. Somatosensation – Pain

Support: NINDS R01NS109936

Title: Gs-coupled receptor signaling through rapgef2 contributes to nociceptor sensitivity and inflammatory hyperalgesia

Authors: *Z. ABBASI[1,2], D. C. MOLLIVER[3];
[1]UNIVERSITY OF NEW ENGLAND, BIDDEFORD, ME; [2]Grad. Sch. of Biomed. Sci. and Engin., Univ. of Maine, Orono, ME; [3]Univ. of New England, Univ. of New England, Biddeford, ME

Abstract: Members of the Rap guanine nucleotide exchange factor (Rapgef) family activate Rap/Ras signaling pathways by exchanging GTP for Rap-bound GDP. Four of the 6 Rapgefs have been identified as effectors of cAMP: Rapgef2/Pdz-gef1, Rapgef3/Epac1, Rapgef4/Epac2, and Rapgef6/Pdz-gef2. Protein kinase C (PKC)/ extracellular signal-regulated kinase (ERK) signaling are required for some components of nociceptor sensitization, therefore Rapgef isoforms may be important effectors in the induction of inflammatory hyperalgesia by Gs-coupled receptors (GsPCRs). We recently identified a role for Epac2-dependent PKC activation in acute nociceptor sensitization and heat hyperalgesia evoked by prostaglandin E2 (PGE2). However, studies in heterologous systems suggest that activation of PKC/ERK downstream of GsPCRs is mediated by Rapgef2. To examine the relative contributions of Rapgef family members to GsPCR signaling in sensory neurons of the dorsal root ganglia (DRG), we first measured the relative expression of Rapgef family members in DRG from male C57BL/6J mice by qPCR. All six Rapgef family members are differentially expressed in DRG, consistent with published mouse and human DRG single cell RNA-seq data indicating that Rapgef2 and

Rapgef4 are the most highly expressed Rapgefs in nociceptive sensory neurons. Immunohistochemistry revealed Rapgef2 immunoreactivity in most or all DRG neurons but not in non-neuronal cells. Deletion of the cAMP-binding site of Rapgef2 in Trpv1-lineage mouse DRG neurons led to a sex-dependent deficit in baseline behavioral thresholds to noxious heat but not cold or mechanical stimuli, and in acute hyperalgesia evoked by Beraprost, an agonist for the Gs-coupled prostacyclin receptor (Ptgir), which by RNA-seq is highly expressed GsPCR in Trpv1-expressing neurons in both human and mouse. These findings provide evidence that cAMP signaling through Rapgef2 contributes to nociceptor sensitivity to noxious heat, but not to mechanical or cold stimuli, despite widespread expression of Rapgef2 in DRG neurons.

Disclosures: Z. Abbasi: None. **D.C. Molliver:** None.

Poster

PSTR142. Inflammatory Pain

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR142.15/V7

Topic: D.02. Somatosensation – Pain

Support: R01NS128403

Title: Calcium release activated calcium (CRAC) channel Orai1 is indispensable for P2X7R-mediated cytokine production

Authors: *V. RAI[1], H. BIRLA[1], X. GAO[2], F. WANG[3], Y. TAO[4], H. HU[5];
[1]Anesthesiol., Rutgers, The State Univ. of NJ - Newark, Newark, NJ; [2]Pharmacol. and Physiol., Drexel Univ., Philadelphia, PA; [3]Anesthesiol., Rutgers Univ., Newark, NJ; [4]Anesthesiol., Rutgers New Jersey Med. Sch., Newark, NJ; [5]Anesthesiol., NJMS, Newark, NJ

Abstract: Calcium release activated calcium (CRAC) channel Orai1 is indispensable for P2X7R-mediated cytokine production Vipin Rai, Hareram Birla, Xinghua Gao, Fengying Wang, Yuanxiang Tao, Huijuan Hu.Department of Anesthesiology, Rutgers New Jersey Medical School, Newark, NJ 07103**Abstract**The P2X7 receptor (P2X7R) is a member of the ATP-gated cationic channel family of P2X.P2X7R plays an important role in chronic pain associated with inflammation and nerve injury.However, the mechanisms underlying the involvement of P2X7R in chronic pain remain unclear.Orai1, the calcium release activated calcium (CRAC) channel subunit, has recently emerged as awidespread pathway for regulating many calcium dependent functions. Here we show that P2X7Ractivation with ATP or the specific P2X7R agonist 2′(3′)-O-(4-Benzoylbenzoyl) adenosine 5′-triphosphate (BzATP) stimulates the production of IL-6 in cultured spinal astrocytes, which isimpeded by a specific P2X7 inhibitor. Interestingly, Orai1 inhibition, knockdown or knockoutdrastically reduces P2X7-induced IL-6 production, while knockdown of STIM1 or STIM2 has nosuch effect, suggesting a store-independent mechanism. To understand how Orai1 is involved inP2X7-mediated cytokine production, we performed calcium imaging recordings. Calcium imagingresults demonstrate that activation of P2X7 with

BzATP leads to calcium release and entry.Western blot analysis reveals that ERK, CaMKII, JNK, and p38 are increased after activation ofP2X7. Inhibition of ERK, CaMK II, and JNK decreases P2X7-mediated IL-6 production inastrocytes. Orai1 knockdown or deficiency markedly reduces the BzATP-induced activation ofCaMKIIα, ERK, and JNK. To determine the role of astrocytic Orai1 in P2X7R-relatedinflammatory pain, we generated conditional Orai1 knockout mice by crossing Orai1fl/fl malemice with female GFAP-Cre mice. Using complete Freund's adjuvant (CFA)-induced chronicinflammation pain model, von Frey and Hargreaves tests, we found that mechanical and thermalhypersensitivity is significantly reduced after two weeks of CFA injection in Orai1 conditional KOmice. Consistently, Western blot results show that deletion of astrocytic Orai1 reduces CFA-induced the increase of IL-6 and TNF-α. These results demonstrate a functional link between Orai1and P2X7R in astrocyte cytokine production. This study provides novel insights into P2X7-mediated inflammatory pain and could open a new avenue to treat inflammatory diseases.Keywords: Astrocyte, purinergic receptor P2X7 (P2X7R), store-operated calcium channels, Orai1,CaMKII, ERK, BzATP, pain, complete Freund's adjuvant (CFA).

Disclosures: V. Rai: None. **H. Birla:** None. **X. Gao:** None. **F. Wang:** None. **Y. Tao:** None. **H. Hu:** None.

Poster

PSTR142. Inflammatory Pain

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR142.16/V8

Topic: D.02. Somatosensation – Pain

Support: CMNDMC11207
NSTC 111-2314-B-384-012 -

Title: Microrna-29a participates in the regulation of inflammatory pain by regulating the expression of interferon-α/ß receptor and mitogen activated protein kinase pathway

Authors: *P.-H. TAN[1,3], K.-F. WANG[2], T.-C. CHIU[2], J.-Y. CHEN[2];
[2]Anesthesiol., [1]Chi-Mei Med. Ctr., Tainan, Taiwan; [3]Sch. of Med., Natl. Sun Yat-Sen Univ., Kaohsiung, Taiwan

Abstract: Interferons (IFNs), such as type-I IFN (IFN-α and ß) and type-II IFN (IFN-γ) were known to be produced by immune cells to elicit antiviral effects. IFNs are also produced by glial cells in the CNS to regulate brain functions. As a proinflammatory cytokine, IFN-γ drives neuropathic pain by inducing microglial activation in the spinal cord. IFN- ß have been demonstrated to exert a direct protective effect against neurotoxic and inflammatory insults on neurons. However, little is known about the role of IFN- ß in regulating pain sensitivity and synaptic transmission in the spinal cord. In our previous microRNA (mir) microarray data under inflammatory pain, a significant increase of mir 29a was found. IFN-α/ ß receptor was known to

be targeted by mir 29a. Thus, we will study the role and mechanism of IFN-ß and mir 29a in regulating inflammatory pain. Intradermal injection of complete Freund's adjuvant (CFA) was performed to induce inflammatory pain. IFN-β, IFN α/ ß receptor, and mir29a in the spinal cord were analyzed on the 1st, 3rd, 5th, 7th, and 10th day after injection of CFA. To investigate the effect of IFN-β and mir 29a on inflammatory pain, IFN-β, and mir 29a mimics were injected intrathecally on 1st day after injection of CFA. Mir 29a inhibitor was injected intrathecally on the 7th day after injection of CFA. After the behavioral test, spinal cords were harvested for analysis of mir 29a, IFNα/ ß receptor, interferon-stimulating gene 15 (ISG15), and p-ERK. Mir 29a was significantly upregulated 3 days after injection of CFA. IFN-α/ ß receptors were significantly downregulated 3 days after injection of CFA. IFN-β were progressively upregulated from 1st day after injection of CFA. Intrathecal injection of mir 29a inhibitor on the 7th day after injection of CFA could significantly increase the mechanical threshold and increased the expression of IFN-α/ ß receptor and ISG15. Simultaneously, the expression of p-ERK was decreased. Intrathecal injection of mir 29a mimic on 1st day after injection of CFA could inhibit the increase of mechanical withdrawal threshold induced by IFN-β. At the same time, the downregulation of IFN-α/ ß receptor, ISG15, and upregulation of p-ERK was noted after injection of mir 29a mimic. Intrathecal injection of 100 ng mir 29a mimic in naïve rats could decrease the paw mechanical withdrawal threshold. Mir 29a was colocalized with IFN-α/ ß receptor. In conclusion, IFN-β and IFN-α/ ß receptors participate in the regulation of CFA-induced pain and mir 29a could regulate the CFA-induced pain by inhibiting the expression of IFN-α/ ß receptor, ISG 15, and activating p-ERK.

Disclosures: P. Tan: None. **K. Wang:** None. **T. Chiu:** None. **J. Chen:** None.

Poster

PSTR142. Inflammatory Pain

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR142.17/V9

Topic: D.02. Somatosensation – Pain

Support: National Science and Engineering Research Council (NSERC)
Quebec Pain Research Network (FRQS-QPRN)
The Louise and Alan Edwards Foundation (LAEF)
Canadian Institutes of Health Research (CIHR)

Title: Evaluating the Impact of Viral TRESK Overexpression on Inflammatory Pain

Authors: *J. SCHWEIZER[1], P. A. SEGUELA[2];
[1]McGill Univ. Integrated Program in Neurosci., Montreal, QC, Canada; [2]Montreal Neurolog. Inst., Montreal, QC, Canada

Abstract: Chronic pain affects around 20% of the world population. Yet a reliable treatment for its management still eludes us. In many cases, chronic pain is a result of peripheral sensitization,

where nociceptive C-fibers from dorsal root ganglia (DRGs) undergo gene expression changes resulting in hyperexcitability and ectopic firing. Among C-fibers, the Transient Potential Vanilloid Receptor 1 (TRPV1)-expressing neurons respond to capsaicin and are involved in thermal and inflammatory pain. As a result, these TRPV1+ nociceptors are prime targets for gene therapy driven treatments of inflammatory pain. If we can reliably target them with a viral vector, we may be able to exploit viral transducing properties to reverse nociceptor hyperexcitability and reduce inflammatory pain in patients. Using a Cre-dependent adeno-associated virus (AAV) in a TRPV1-Cre mouse line, we aim to reverse the hyperexcitability in inflammatory pain by overexpressing TWIK-related spinal cord potassium (TRESK) channel. Typical TRESK currents were recorded in transfected HEK cells expressing rodent or human TRESK. We validated the inhibitory effects of loratadine and Ca^{2+}-mediated potentiation on human TRESK currents. We also created a human TRESK-HA construct which does not impact channel function. The same construct was subcloned into a pAAV-CAG-DIO plasmid to produce an AAV-CAG-DIO-hTRESK-HA used in a TRPV1-Cre mouse line. Through intraperitoneal co-injection of AAV-CAG-DIO-hTRESK-HA and AAV-CAG-DIO-mCherry in P0-P5 pups, we are assessing the effect of TRESK over-expression on naïve and CFA animals. Animals were tested behaviorally 4 weeks post-injection on the Hargreaves and von Frey apparatus to assess normal responses to thermal and mechanical stimuli. Half of the animals were then injected with intraplantar CFA and reassessed in sensory assays for two weeks post injection. DRGs were collected from the other half of the animals for immunohistochemistry (IHC) and electrophysiological recordings in primary cultures. The same procedure was followed for CFA-injected animals following behavioral tests. Our preliminary results seem to indicate that TRPV1+ neurons from DRG cultures of animals injected with AAV-CAG-DIO-hTRESK-HA display an increased rheobase and are hypoexcitable compared to control neurons. We are currently in the process of collecting behavioral and electrophysiological data. Now that we have validated our AAV-CAG-DIO-hTRESK-HA construct, we have moved on to characterizing the effect of TRESK over-expression in TRPV1+ neurons in vivo. Our preliminary data suggest that this could prove an effective treatment for inflammatory pain.

Disclosures: **J. Schweizer:** None. **P.A. Seguela:** None.

Poster

PSTR142. Inflammatory Pain

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR142.18/V10

Topic: D.02. Somatosensation – Pain

Support: NIH Grant NS120486

Title: Adaptations of the endocannabinoid system during persistent inflammation are driven by corticosterone within the vlPAG

Authors: *B. COUTENS[1,2], K. B. MCPHERSON[2], B. BOSTON[2], C. BOUCHET[3], S. L. INGRAM[2];
[1]Univ. of Colorado, Anschutz Med. Neurosci. Grad. Training Program, Aurora, CO; [2]Univ. of Colorado, Anschutz Med. Campus, Aurora, CO; [3]Colorado State Univ., Colorado State Univ., Fort Collins, CO

Abstract: Chronic pain exerts an enormous personal and economic burden, affecting more than 30% of people worldwide and the treatment of this disorder in the clinic remains a major challenge. The ventrolateral periaqueductal gray (vlPAG) integrates inputs from many brain regions associated with the processing of nociceptive, cognitive, and affective components of chronic pain perception, and is a key brain area for opioid and endocannabinoid modulation of pain. Both opioids and endocannabinoids (eCBs) decrease GABA release through their presynaptic receptors (MORs and CB1Rs, respectively). Chronic inflammation induced by injections of Complete Freud's adjuvant (CFA) into a hindpaw desensitizes CB1Rs while sensitizing MOR responses. Interestingly, the development of CB1R desensitization appears to be faster in females than in males, indicating a sex difference in the regulation of the endocannabinoid system. The current studies test the hypothesis that CFA-induced inflammation increases corticosterone levels that mediate the observed adaptations in CB1Rs and MORs in the vlPAG of male and female rats. Using whole-cell patch-clamp recordings from neurons in *ex vivo* slices containing the vlPAG from Sprague-Dawley male and female rats, we first observed that activation of the glucocorticoid receptor (GR) by corticosterone rapidly increases and prolongs eCB tone resulting in inhibition of presynaptic GABA release via activation of CB1Rs. These effects are abolished by inhibition of 2-arachidonoylglycerol (2-AG) synthesis. The rapid effects suggest that corticosterone does not act through nuclear receptors but through membrane receptors. Of note, the effects of corticosterone are independent of MORs. In CFA-treated rats, despite CB1R desensitization, depolarization-induced suppression of inhibition (DSI)-induced eCB release is prolonged and dependent on monoacylglycerol lipase (MAGL) activity. Future studies will examine the role of nuclear GR in regulating MAGL levels. Taken together our result show that eCB system is regulated by corticosterone which could act through both nuclear and membrane receptors in a time-dependent manner.

Disclosures: B. Coutens: None. **K.B. McPherson:** None. **B. Boston:** None. **C. Bouchet:** None. **S.L. Ingram:** None.

Poster

PSTR142. Inflammatory Pain

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR142.19/V12

Topic: D.02. Somatosensation – Pain

Title: Endogenous Derivatives of Linoleic Acid and Their Stable Analogues are Potential Pain Mediators

Authors: *J. WHEELER[1], A. F. DOMENICHIELLO[2], G. KEYES[3], K. MAIDEN[3], J. M. DAVIS[4], C. RAMSDEN[3], S. K. MISHRA[5];
[1]North Carolina State Univ., Raleigh, NC; [2]NIA/NIH, NIA/NIH, Washington, DC; [3]NIH, Bethesda, MD; [4]Univ. of Illinois, Chicago, Chicago, IL; [5]Col. of Vet. Medicine, NC State Universit, Col. of Vet. Medicine, NC State Universit, Raleigh, NC

Abstract: Omega 6 (ω-6) fatty acid metabolites are common endogenous mediators of pain and inflammation. Pain mediators derived from n-6 arachidonic acid are well recognized; however, linoleic acid is the most abundant n-6 fatty acid in modern industrialized diets. Despite this dietary prevalence, several oxidized linoleic acid metabolites (OxLAMs) have been mechanistically linked to pathological conditions ranging from cardiovascular disease to chronic pain. Rodents fed a diet enriched in linoleic acid to mimic the modern western diet had an increased concentration of OxLAMs in tissues associated with idiopathic pain syndromes. In humans, migraine and chronic headaches are common idiopathic pain syndromes. In clinical trials, decreasing the amount of dietary ω-6 fatty acids resulted in a decrease in the number of headache days per month. Further, several OxLAMs were found to have decreased concentrations in headache patients with this decreased ω-6 fatty acid diet. Here, we characterize the functional role of one of these mediators, 13-hydroxy-9,10-epoxyoctadecenoate, and its metabolite, 13, 9, 10-trihydroxyoctadecenoate. Using calcium imaging, we first determined that these two compounds are capable of activating sensory neurons. Next, we determined, using the subcutaneous cheek and intraplantar injection, that these OxLAMs induce pain behaviors and thermal hypersensitivity. Additionally, we used these *in vitro* and *in vivo* assays to determine the potentially active pharmacophore of these OxLAMs. Taken together, our results demonstrate a role in pain for two OxLAMs associated with migraine and chronic headaches and/or cutaneous pain associated with psoriasis.

Disclosures: J. Wheeler: None. **A.F. Domenichiello:** None. **G. Keyes:** None. **K. Maiden:** None. **J.M. Davis:** None. **C. Ramsden:** None. **S.K. Mishra:** None.

Poster

PSTR142. Inflammatory Pain

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR142.20/Web Only

Topic: D.02. Somatosensation – Pain

Title: Anti-inflammatory effect of Taxodium huegelli in a model of intraplantar edema

Authors: L. ONOFRE-HURTADO, L. MACÍAS-ROSALES, M. TINOCO-MÉNDEZ, M. RIVERA-HUERTA, *A. CARBALLO;
UNAM, Coyoacán, Mexico

Abstract: Inflammation is one of the most common problems that afflict people, being a call for attention or alert of the organism; however, its expansion causes damage to cells and tissues, so it

would be convenient to study new alternatives or solutions that reduce or eradicate it without producing adverse effects. Due to this, currently the treatments seek to reduce the risks by enhancing the benefits and within them we find therapeutic alternatives such as natural products; Among these, the use of plants as an alternative or adjuvant in treatments stands out; Such is the case of Taxodium huegelli (ahuehuete).
Goals: To evaluate the anti-inflammatory effect of the ethanolic extract of ahuehuete in the intraplantar edema model.
Methods: Male Wistar rats between 180 g and 200 g were used in groups of 6 animals per treatment, the anti-inflammatory effect of 10 mg/kg, 100 mg/kg, 316.2 mg/kg and 562.3 mg/ kg with the dose of meloxicam of 10 mg/kg (reference drug) the model of intraplantar edema induced by intraplantar injection at 1% was used; The degree of inflammation was evaluated by measuring the displaced volume of the hind leg of the animal with the help of the plethysmometer equipment (IITC Life Science), the data obtained were analyzed with the SPSS V.25 program.
Results and discussion: The ahuehuete extract produces an anti-inflammatory effect in the plantar edema model by significantly reducing the degree of inflammation with respect to its vehicle. Compared to the reference drug meloxicam (10 mg/kg), ahuehuete concentrations of 10 mg/kg and 316.2 mg/kg demonstrated a similar anti-inflammatory effect, while the concentration of 562.3 mg/kg produced a anti-inflammatory effect significantly higher than that of meloxicam.
Conclusions: The extracts of ahuehuete produce an anti-inflammatory effect in the model of plantar edema in rats statistically similar to the reference drug (meloxicam).

Disclosures: L. Onofre-Hurtado: None. **L. Macías-Rosales:** None. **M. Tinoco-Méndez:** None. **M. Rivera-Huerta:** None. **A. Carballo:** None.

Poster

PSTR142. Inflammatory Pain

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR142.21/V13

Topic: D.02. Somatosensation – Pain

Support: PAPIIT IN201723
CONCAYT 319379

Title: Quercetin modifies the antinociceptive and ulcerogenic effect of indomethacin but not its anti-inflammatory effect in rats

Authors: *J. AVILES-HERRERA[1], G. E. ANGELES-LOPEZ[1], R. VENTURA-MARTÍNEZ[1], M. DECIGA-CAMPOS[2];
[1]Dept. of Pharmacol., Univ. Nacional Autonoma de Mexico, Ciudad de México, Mexico;
[2]Sección de Estudios de Posgrado e Investigación, Inst. Politécnico Nacional, Ciudad de México, Mexico

Abstract: Diseases that affect the musculoskeletal system are considered one of the main causes of chronic pain and the non-steroidal anti-inflammatory drugs (NSAIDs) are commonly used for its treatment. However, NSAIDs do not always produced the expected analgesic efficacy, so their lack of effectiveness has motivated the search for other alternatives such as the combination of drugs. On the other hand, quercetin (a bioflavonoid) has shown antinociceptive effects in several pain models, hence, the purpose of this work was to evaluate the pharmacological interaction of quercetin on the antinociceptive, anti-inflammatory, and ulcerogenic effects of indomethacin on rats. To induce nociception, 50 µl of 30% uric acid was administered in the right posterior knee of each rat. The uric acid-induced dysfunction in the animals were recorded until the animals showed a complete dysfunction. The recovery of their functionality after administration of indomethacin (10 mg/kg, p.o.) alone or with quercetin (100 mg/kg, i.p.) was interpreted as antinociceptive effect. In other group of animals, the anti-inflammatory effect of both treatments was evaluated using the carrageenan model, in which the induced edema by the intraplantar administration of 100 µl of 1% carrageenan was measured. Finally, the gastric damaged induced by indomethacin alone and after 10-days of treatment with quercetin was also evaluated. The results shows that quercetin decreased the antinociceptive effect of indomethacin from 50% to 28% (49.24 $\pm$ 4.61 vs 27.9 $\pm$ 7.33), but it did not modify the anti-inflammatory effect of indomethacin (21.93 $\pm$ 4.17 vs 24.59 $\pm$ 5.51 % edema). Also, the macroscopic analysis of the stomachs showed that indomethacin produced an area of injury of 39.7 $\pm$ 10.7 cm^2, while indomethacin with quercetin result in a larger area of injury of 61.5 $\pm$ 17.9 cm^2. In conclusion, even though quercetin did not modify the anti-inflammatory effect of indomethacin, it significantly decreased its antinociceptive effect and increased its ulcerogenic effect. These results suggest that indomethacin should not be administered with quercetin because it negatively modifies its pharmacological effects.

Disclosures: J. Aviles-Herrera: None. **G.E. Angeles-Lopez:** None. **R. Ventura-Martínez:** None. **M. Deciga-Campos:** None.

Poster

PSTR143. Olfaction and Gustation

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR143.01/V14

Topic: D.04. The Chemical Senses

Title: Structure and Functional Impact of Neural Circuits Mediating Crosstalk among Gustatory Receptor Neurons (GRNs) in D. melanogaster

Authors: *B.-M. SONG, Z. ALDWORTH, M. A. STOPFER;
NIH, Bethesda, MD

Abstract: A healthy sense of taste is essential for quality of life. Understanding the process of gustatory coding will provide insight into how our brain works as well as how gustatory pathologies arise. Tasting starts with the activation of sensory detectors, Taste Receptor Cells

(TRCs) in mammals and Gustatory Receptor Neurons (GRNs) in insects. Conventionally, the function of these sensory detectors is thought to be limited to simply responding to gustatory cues and transmitting the resulting signals to the brain. It is also thought that gustatory information is transmitted mainly through a small number of separate channels, each dedicated to a different taste. However, several lines of evidence in both mammals and insects suggest that taste detectors play an additional role in gustatory coding by regulating each other's activity. Yet, the gustatory circuitry that mediates such crosstalk and its functional role in gustatory coding is poorly understood.Here, in the fruit fly *D. melanogaster*, we report a novel type of gustatory circuitry at single cell resolution. We employed an anatomical screen using Trans-Tango, an anterograde transsynaptic mapping tool, and identified five candidate GRNs that are synaptically interconnected, potentially forming 20 GRN pairs. Using electrophysiology, we demonstrated that, in four of the GRN pairs, directly activating each of these presynaptic GRNs significantly increased spiking in their putative postsynaptic GRNs. Next, with specific genetic manipulations combined with electrophysiology, we determined the unique identities of each presynaptic and postsynaptic GRN as well as the functional impact of crosstalk between them. We further identified the neurotransmitter receptor expressed by the postsynaptic GRNs. In the mutant flies lacking this receptor, crosstalk between the 4 GRN pairs was either abolished or substantially diminished. To determine the functional role of crosstalk between GRNs, we are now testing gustatory behavior in the null mutant flies. Our discovery highlights that a change in a single taste detector is sufficient to change the activity of its follower taste detectors, emphasizing the importance of understanding how a population of interacting taste detectors achieves gustatory coding.

Disclosures: **B. Song:** A. Employment/Salary (full or part-time); Eunice Kennedy Shriver National Institutes of Child Health and Human Development, Section on Sensory Coding and Neural Ensembles, NIH, Bethesda, MD, USA. **Z. Aldworth:** A. Employment/Salary (full or part-time); Eunice Kennedy Shriver National Institutes of Child Health and Human Development, Section on Sensory Coding and Neural Ensembles, NIH, Bethesda, MD, USA. **M.A. Stopfer:** A. Employment/Salary (full or part-time); Eunice Kennedy Shriver National Institutes of Child Health and Human Development, Section on Sensory Coding and Neural Ensembles, NIH, Bethesda, MD, USA.

Poster

PSTR143. Olfaction and Gustation

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR143.02/V15

Topic: D.04. The Chemical Senses

Support: NIH Grant P20GM125508
Hawaii Community Foundation 18CON-90818
NIH Grant R24OD030214

Title: Higher olfactory sensing may evolve through more motile cilia at the olfactory epithelium but not the increase of the olfactory receptor genes in the blind cavefish.

Authors: *N. CHOI[1], M. NIKAIDO[2], E. RICE[3], W. WARREN[3], M. YOSHIZAWA[1]; [1]Univ. of Hawaii, Honolulu, HI; [2]Tokyo Inst. of Technol., Yokohama, Japan; [3]Bond Life Sci. Ctr., Univ. of Missouri, Columbia, MO

Abstract: Typified in the super-sensing ability of sharks for finding prey and salmons for homing, olfactory sensing plays a crucial role in adaptation. The diversity of olfactory receptors (OR), which is a part of the superfamily of the 7-transmembrane G-coupled receptors, has attracted strong interest from the beginning of evolutionary studies. Dr. Masatoshi Nei studied OR evolution in vertebrates and proposed that the elaboration of visual sensing has an antagonistic relationship with OR diversification—loss of functional OR genes—, as seen in primates (e.g., monkeys) compared to the other mammalian lineages (e.g., dogs). To test whether this antagonistic evolution between vision and olfactory sensing is a general rule, we used a powerful evolutionary model, the Mexican tetra. The cave-dwelling population lost the eyes and gained 10^5 times higher amino acid sensitivity compared to its sighted surface-dwelling population (10^{-10} M vs 10^{-5} M of alanine in the behavioral response, respectively). In this study, we first compared the deeply sequenced high-quality genomes (combination of PacBio, Hi-C and ATAC-seq) of 1 sighted surface and 4 independently-evolved blind cave populations by identifying the functional ORs: Vasopressin receptor 2 (V2Rs), Taste receptor type 2 (T2Rs), and Trace amino acid receptors (TAARs). Surface fish have 159 ORs, 52 V2R, 25 T2R, and 77 TAAR, which are slightly smaller than but comparable to typical in teleosts. Surprisingly, we found slightly smaller numbers of ORs (142-148 ORs) and other receptor families in four blind cave populations, which not only contradicted our prediction of the antagonistic evolution between vision and olfactory sensors but also did not explain the 10^5 times higher olfactory sensing observed in cavefish. We then sought morphological elaborations. The scanning and transmission electron microscopy revealed extensive amounts of motile cilia in a cave population's olfactory epithelium. We will present the most updated mapping result on olfactory receptors and the evolution of olfactory sensation in compensation for the loss of vision.

Disclosures: **N. Choi:** None. **M. Nikaido:** None. **E. Rice:** None. **W. Warren:** None. **M. Yoshizawa:** None.

Poster

PSTR143. Olfaction and Gustation

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR143.03/V16

Topic: D.04. The Chemical Senses

Support: NIDCD Grant DC019636

Title: Diverse determinants of the temporal dynamics of odorant representations by glomeruli of the mouse olfactory bulb

Authors: *E. ACQUAH, S. SHORT, M. WACHOWIAK;
Neurobio., Univ. of Utah, Salt Lake City, UT

Abstract: Diverse determinants of the temporal dynamics of odorant representations by glomeruli of the mouse olfactory bulb Elvis Acquah, Shaina Short, Matt Wachowiak.In the mammalian olfactory system, the temporal dynamics of odorant-evoked activity plays an important role in encoding odor information, yet the determinants of distinct temporal patterns remain unclear. The 'primacy' model hypothesizes that inhalation-linked response latencies reflect relative sensitivity to an odorant, with the most sensitive neurons responding earliest. We tested this model by defining relative sensitivities of glomeruli in the mouse olfactory bulb (OB) to specific odorants and characterizing their response dynamics across concentrations. We also tested alternate models by relating responses to odorants with diverse chemical structures. We used two-photon imaging in awake, head-fixed mice expressing genetically encoded calcium or glutamate reporters in olfactory sensory neurons (OSNs) or mitral/tufted (MT) cells, and built on recent work establishing 'primary' glomeruli for particular odorants (Burton et al., doi: 10.7554/eLife.80470). Increasing odorant concentration recruited activation in 'non-primary' glomeruli, but initial response latencies and subsequent inhalation-linked dynamics only sometimes correlated with relative sensitivity, inconsistent with the primacy model. However, temporal dynamics were strongly predicted by a combination of odorant chemical features and glomerular location. In particular, glomeruli in the class I OSN domain showed faster responses to acids than to their corresponding esters and aldehydes, while ester-sensitive glomeruli in the class II domain were less-sensitive but responded more rapidly. These results are consistent with rapid (i.e., sub-sniff) conversion of odorants by xenobiotic enzymes known to be present in the nasal epithelium. These relationships persisted across concentrations and were apparent among MT cell glomerular signals as well as OSN responses, suggesting that delayed activation of OSN input drives delayed activation, rather than suppression, of MT cells. Overall, these results suggest that relative response latencies do not robustly reflect relative sensitivity to an odorant, and that factors such as odorant chemistry, perireceptor processes, and glomerular identity may be even stronger determinants of the dynamics of glomerular odor representations.

Disclosures: E. Acquah: None. **S. Short:** None. **M. Wachowiak:** None.

Poster

PSTR143. Olfaction and Gustation

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR143.04/V17

Topic: D.04. The Chemical Senses

Support: Life Science Research Foundation
NIH
HHMI

Title: The organization of the early olfactory system in the clonal raider ant.

Authors: D. D. FRANK, D. J. C. KRONAUER;
Rockefeller Univ., New York, NY

Abstract: Human neurobiology has evolved to support advanced social cognition, cooperation and communication. These phenomena enable human sociality, yet we know little of their foundation in the brain, and neuroscience studies in genetic model organisms have primarily investigated ubiquitous social behaviors like aggression and mating. The clonal raider ant *Ooceraea biroi* is a distinctly effective model organism to fill this gap: ants form complex societies not found in conventional model organisms and the experimental accessibility of the clonal raider ant provides a powerful platform unavailable to studies involving highly social mammals.
My research uses state-of-the-art neurogenetic tools to study the cellular basis of chemical communication in the clonal raider ant nervous system. Ants have a remarkable capacity for chemical communication. Information is encoded by large arrays of pheromones exuded by dedicated exocrine glands and is received and processed by highly advanced olfactory systems. With approximately 500 olfactory glomeruli, the clonal raider ant antennal lobe (AL) is more complex than other insects and is evocative of the olfactory bulb in the brain of mammals (*Drosophila* have ~50 AL glomeruli, for reference). Recent studies in our lab suggests the evolution of sociality may have coincided with unique neurodevelopmental logic in the ant antennal lobe and that, surprisingly, its similarity to the mammalian olfactory bulb may include developmental mechanisms. Here, I use transgenic ants, in vivo two-photon GCaMP imaging, and confocal microscopy to investigate the organization of the olfactory system in the clonal raider ant.

Disclosures: D.D. Frank: None. **D.J.C. Kronauer:** None.

Poster

PSTR143. Olfaction and Gustation

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR143.05/Web Only

Topic: D.04. The Chemical Senses

Support: SB/SJF/2021-22/04-C

Title: Response plasticity in an olfactory sensory neuron on the maxillary palps of Aedes aegypti

Authors: *S. SARAN SINGH[1], S. GARG[2], P. SINGH[2], S. GUPTA[2], A. AIRAN[2], S. GOYAL[2], N. GUPTA[2];
[1]Indian Inst. of Technology, Kanpur, Gorakhpur, India; [2]Indian Inst. of Technol., Kanpur, India

Abstract: The mosquito *Aedes aegypti* spreads various deadly diseases like dengue, zika fever, yellow fever, and chikungunya across the globe. Female mosquitoes detect human hosts using a variety of cues including olfactory signals such as skin odor and carbon dioxide. One of the main olfactory organs, the maxillary palp, detects these odorants through several small sensilla present on their surface. Each of these sensilla, known as capitate pegs, houses three sensory neurons. The three neurons are named according to their spike sizes: cpA (largest), cpB (mid-sized), and cpC (smallest). We have studied the capitate peg sensillum neurons and their characteristics. We noticed an interesting characteristic of the cpC neuron's activity while recording. The capitate peg sensillum consistently showed a large deflection in the local field potential in the first trial of testing with the odorant 1-octen-3-ol, and these parameters gradually decreased over the subsequent trials. A similar plasticity in the LFP response was also observed for other odorants. We then checked the involvement of cpA, cpB, and cpC neurons in the plasticity of the LFP response. We found that the plasticity in the LFP originates from an analogous plasticity in the firing rate of the CpC neuron: this neuron showed reducing firing rates to successive presentations of the stimulus. This reduction in responses was specific to the cpC neuron, as it was not observed in cpA or cpB neurons. We finally checked if there was a functional significance to the response plasticity by checking the activity of the downstream neurons, and found that the plasticity observed in the sensory neurons affects the responses of the downstream projection neurons. These results contribute to an understanding of the olfactory processing in the maxillary palps of *Aedes aegypti*.

Disclosures: S. Saran Singh: None. **S. Garg:** None. **P. Singh:** None. **S. Gupta:** None. **A. Airan:** None. **S. Goyal:** None. **N. Gupta:** None.

Poster

PSTR143. Olfaction and Gustation

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR143.06/V18

Topic: D.04. The Chemical Senses

Title: Differences in sex-steroid metabolism in olfactory and vomeronasal sensory cells

Authors: *S. TAKAMI[1], S. HORIE[2];
[1]Dept. of Physical Therapy, Japanese Sch. of Technol. for Social Med., Tokyo, Japan; [2]Dept. of Anat., Kawasaki Med. Sch., Okayama, Japan

Abstract: Sensory cells of the main olfactory and vomeronasal system (MOS and VNS, respectively), olfactory and vomeronasal sensory cells (OSCs and VSCs) share cellular properties of epithelial cells and neurons. Immature OSCs and VSCs differentiate from

progenitor cells that are present in the olfactory and vomeronasal sensory epithelia (OE and VSE), and eventually become mature bipolar neurons that make synaptic contacts with the primary brain centers. Although androgens and estrogens, which are referred to as sex steroids, are known to make several effects on the MOS and VNS, little was known about cellular mechanism about their metabolism in the OE and VSE. To obtain basic knowledge about functional roles of sex steroids in the OE and VSE, we examined rat OSCs, VSCs, and their sustentacular (Sus) cells of adult Sprague-Dawley rats using immunocytochemical techniques. In the OE, immunoreactivity (IR) for steroid side chain-cleaving enzyme (P450scc), 17beta-hydroxysteroid dehydrogenase type 1 (17 beta-HSD-1), and 17 beta-HSD type 2 (17 beta-HSD-2) were localized in olfactory Sus cells, whereas no IR in OSCs. In the VSE, however, IR for 17 beta-HSD-1 and -2 were localized in VSCs, whereas no IR for P450scc in these cells. By contrast, no IR for the above three enzymes was detected in vomeronasal Sus cells. In addition, both OSCs and VSCs contain IR for estrogen receptors; beta-type (ER beta) in the OSCs and alpha-type (ER alpha) in the VSCs. Immunoelectron microscopy demonstrated that IR for 17 beta-HSD-1 and -2 were localized in smooth endoplasmic reticulum of olfactory Sus cells and VSCs. These results suggest that (1) OSCs take up estrogens from olfactory Sus cells via ER beta and (2) VSCs metabolize estrogens and/or androgens using 17beta-HSD-1 and -2 produced by themselves, and also take up estrogens from intraepithelial capillaries via ER alpha. It is also possible that sex steroids metabolized within VSCs are transported via axonal flow to the primary brain center. OSCs and VSCs, which are bipolar neurons, may receive similar effects by estradiol as demonstrated in the central nervous system.

Disclosures: S. Takami: None. **S. Horie:** None.

Poster

PSTR143. Olfaction and Gustation

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR143.07/V19

Topic: D.04. The Chemical Senses

Support: Intramural funds at TIFR Hyderabad from the Department of Atomic Energy (DAE), India, under Project ID RTI 4007.

Title: Single cell transcriptome of the mouse vomeronasal neuroepithelium indicates a specialized endoplasmic reticulum environment in neuronal subsets.

Authors: *D. G V S, A. DANI;
Tata Inst. of Fundamental Res., Hyderabad, India

Abstract: The vertebrate vomeronasal (VNO) system is an important model to understand how sensory information leads to innate and social behaviors. With the expression of several evolutionarily and functionally distinct genes, VNO neurons offer an intricate system to understand the cellular and molecular biology associated with sensory signaling. We performed

single cell RNA sequencing of the mouse VNO neuroepithelium to understand the diversity, gene co-expression patterns, and functional differences amongst neuronal subtypes. In addition to the known major neuronal subtypes, our analysis reveals a diversity of cell types comprising glia, immune and sustentacular cells, with genes specific to each. Pseudotime developmental analysis indicates that neurons that originate from common progenitors show divergent gene expression during maturation with transient and persistent changes in transcription factor expression at critical branch points. Within mature sensory neurons, we find significantly higher expression of endoplasmic reticulum genes in a neuronal subtype, indicative of a putative functional requirement for protein maturation.

Disclosures: D. G v s: None. **A. Dani:** None.

Poster

PSTR143. Olfaction and Gustation

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR143.08/V20

Topic: D.04. The Chemical Senses

Support: NICHD Intramural grant to MS

Title: Olfactory structure and function in newly hatched and adult locusts

Authors: *K. SUN[1,2], S. RAY[3], N. GUPTA[4], M. A. STOPFER[2];
[1]NIH, Bethesda, MD; [2]Section on Sensory Coding and Neural Ensembles, Eunice Kennedy Shriver Natl. Inst. of Child Hlth. and Human Develop., Bethesda, MD; [3]Plaksha Univ., Mohali, India; [4]Indian Inst. of Technol. Kanpur, Indian Inst. of Technol. Kanpur, Kanpur, India

Abstract: The sense of smell is essential for survival in most animal species as it is needed to find food and mates and to avoid toxic substances and predators. In many animals, olfaction is crucial from the moment of birth. Yet, many questions remain about olfactory development. Adult locust *Schistocerca americana* have provided a useful model for understanding olfaction. Here we compared olfactory structure and function in newly hatched, young, and adult locusts. Our results show that olfactory neurons in the hatchlings are smaller than but otherwise morphologically identical to those in adults. Olfactory projection neurons (PNs) are similar in number in hatchlings and adults, have similar numbers of processes radiating from the center to the periphery of the antennal lobe to similar numbers of glomeruli, and send a single branch to the ipsilateral mushroom body calyx and lateral horn. Local interneurons (LNs) in the antennal lobe show similar morphologies in hatchlings and adults. Intracellular and extracellular recordings of PNs reveal similar responses to odorants, including complex sequences of excitation and inhibition, throughout development. Intracellular recordings from LNs in hatchlings and adults reveal similar responses. Electroantennograms used to measure population responses of antennal olfactory receptor neurons (ORNs) showed similar responses in hatchlings and adults to a range of odorants, although the hatchlings have many fewer ORNs. Local field

potential (LFP) recordings from the mushroom body and intracellular recordings from LNs revealed odor-evoked oscillatory neural activity throughout development.
Notably, LFPs oscillations increased in frequency with development. We hypothesized oscillation frequency was determined by intensity of output from the ORN population (Ito et al, 2009), which increases throughout development. We simulated a transition from adult to hatchling numbers of ORNs by gradually trimming segments from the antenna of intact adults while recording odor-evoked LFPs in the mushroom body. Consistent with our hypothesis, reducing numbers of ORNs gradually reduced oscillation frequency.
Overall, our results reveal the hatchling olfactory system is smaller than, but in structure and function, otherwise nearly identical to the adult.

Disclosures: K. Sun: None. **S. Ray:** None. **N. Gupta:** None. **M.A. Stopfer:** None.

Poster

PSTR143. Olfaction and Gustation

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR143.09/V21

Topic: D.04. The Chemical Senses

Support: NIH Grant U19NS112953

Title: A comprehensive molecular map of olfactory bulb glomeruli generated with 3D spatial transcriptomics

Authors: *N. KLIMPERT[1], M. KOLLO[2], D. J. BARRY[3], D. H. BRANN[4], S. R. DATTA[4], A. T. SCHAEFER[2,5], A. FLEISCHMANN[1];
[1]Neurosci., Brown Univ., Providence, RI; [2]Sensory Circuits and Neurotechnology Lab., [3]Crick Advanced Light Microscopy, The Francis Crick Inst., London, United Kingdom; [4]Dept. of Neurobio., Harvard Med. Sch., Boston, MA; [5]Dept. of Neuroscience, Physiol. and Pharmacol., Univ. Col. London, London, United Kingdom

Abstract: Identifying the spatial organization of glomeruli and molecular domains in the olfactory bulb (OB) is necessary to understand the routing of odor information from periphery to higher olfactory areas. A small number of glomeruli have been genetically characterized, and recent studies have inferred positions of many glomeruli through single-cell RNA sequencing and machine learning. Here, we have used spatial transcriptomics with barcoded microarrays to directly measure gene expression, characterizing glomerular position and generating a comprehensive, 3D molecular map of the entire mouse OB. We performed 10X Visium Spatial Gene Expression on 200 consecutive 10µm sections of mouse OB tissue. We aligned the histological reference images and integrated the resulting 3D alignment with gene expression. We then developed a probabilistic model to define glomerular location based on expression of odorant receptor genes. From this comprehensive, 3D molecular map of OB glomeruli, we can define axes of inter- and intra-bulb symmetry, elucidate neighbor relationships between

glomeruli, and reveal previously uncharacterized molecular domains. Moreover, we have generated spatial transcriptomics datasets on smaller regions of OB from replicate animals. From the alignment of these replicate datasets with the complete molecular map, we can characterize the conservation of glomerular position and neighbor relationships across animals. Finally, by integrating our dataset with single-cell RNA sequencing of the olfactory epithelium, we will elucidate the relationship between olfactory sensory neuron molecular profiles and glomerular organization. This comprehensive molecular map will provide a foundation for future functional studies and extend our understanding of the organization through which odor information is processed.

Disclosures: **N. Klimpert:** None. **M. Kollo:** None. **D.J. Barry:** None. **D.H. Brann:** None. **S.R. Datta:** None. **A.T. Schaefer:** None. **A. Fleischmann:** None.

Poster

PSTR143. Olfaction and Gustation

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR143.10/V22

Topic: F.01. Neuroethology

Support: Gruber Science Fellowship
NIH R01 NS116584
NIH R01 DC018570
Richard and Susan Smith Family Award for Excellence in Biomedical Research
Klingenstein-Simons Fellowship Award
Kavli Institute

Title: Balancing sensory inputs: olfactory and thermosensory guided behaviors

Authors: *E. DICKINSON, J. M. JEANNE;
Yale Univ., New Haven, CT

Abstract: Organisms are constantly bombarded with environmental information across multiple sensory streams. Determining which information to retain, which information to combine, and which information to keep separate is a nontrivial task for the brain. However, it is poorly understood how the brain integrates relevant sensory information across modalities. The fruit fly, *Drosophila melanogaster*, offers a unique opportunity to study flexible interactions of two sensory modalities - temperature and olfaction - that interact to optimize an essential behavior: feeding. Flies rely heavily on their olfactory system to locate and select appropriate food sources. In general, flies sample the concentration of attractive food related chemicals that have been volatilized into the air and use changes in these concentration gradients to navigate to their food source. Additionally, as small poikilotherms, they are highly susceptible to environmental changes in temperature while navigating the world. We are investigating when and how

temperature modulates olfactory guided feeding in flies using novel behavioral assays, genetic screening, and physiological approaches. We have found that temperature robustly modulates flies' behavioral attraction to food odors. Specifically, attraction decreases linearly as temperatures move away from the preferred temperature range of Drosophila (~24-26ºC); conversely, attraction increases nonlinearly as temperatures approach the preferred temperature range. Taken together, our results show that flies employ a behavioral strategy which maximizes feeding opportunities in dynamic and fluctuating temperature environments.

Disclosures: E. Dickinson: None. **J.M. Jeanne:** None.

Poster

PSTR144. Olfaction: Peripheral Mechanisms and Behavior

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR144.01/V23

Topic: D.04. The Chemical Senses

Support: German Research Foundation (Ha4466/11-1, Ha4466/20-1 and SFB 936 178316478 to I.L.H.-O)
European Research Council (ERC-2015-CoG 681577 to I.L.H.-O.)
Horizon 2020 DEEPER (101016787 to I.L.H.-O.)
Landesforschungsförderung Hamburg (LFF73 and LFF76 to I.L. H.-O.)

Title: Olfactory bulb activity shapes the development of entorhinal-hippocampal coupling and associated cognitive abilities

Authors: *Y.-N. CHEN, J. K. KOSTKA, S. H. BITZENHOFER, I. L. HANGANU-OPATZ; Inst. of Developmental Neurophysiology, Ctr. of Mol. Neurobio., Univ. Med. Ctr. Hamburg-Eppendorf, Hamburg, Germany

Abstract: The interplay between olfaction and higher cognitive processing has been documented in the adult brain, yet its development is poorly understood. In mice, shortly after birth, endogenous and stimulus-evoked activity in the olfactory bulb (OB) boosts the oscillatory entrainment of downstream lateral entorhinal cortex (LEC) and hippocampus (HP). However, it is unclear whether early OB activity has a long-lasting impact on entorhinal-hippocampal function and cognitive processing. Here, we chemogenetically silenced the synaptic outputs of mitral/tufted cells, the main projection neurons in the OB, during postnatal days 8-10. The transient manipulation leads to a long-lasting reduction of oscillatory coupling and weaker responsiveness to stimuli within the developing entorhinal-hippocampal network accompanied by dendritic sparsification of LEC pyramidal neurons. Moreover, the transient silencing reduces the performance in behavioral tests involving entorhinal-hippocampal circuits later in life. Thus, the early OB activity is critical for the functional LEC-HP development and maturation of cognitive abilities.

Disclosures: **Y. Chen:** None. **J.K. Kostka:** None. **S.H. Bitzenhofer:** None. **I.L. Hanganu-Opatz:** None.

Poster

PSTR144. Olfaction: Peripheral Mechanisms and Behavior

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR144.02/V24

Topic: D.04. The Chemical Senses

Support: NICHD Intramural grant to MS

Title: The palp olfactory system utilizes odor codes different from the antennal olfactory system in the locust

Authors: *Z. ALDWORTH, M. STOPFER;
NIH-NICHD, Bethesda, MD

Abstract: Olfaction in insects has proved to be a useful model for studying sensory coding. The relative simplicity and accessibility of the insect brain have been leveraged to examine phenomena that also occur in the mammalian central nervous system, such as synchronous oscillations, ensemble temporal coding, and sparse coding. However, while the vast majority of this research has focused on the pathway extending from the insect antenna to the mushroom bodies and lateral horn in the brain, most insects have secondary olfactory organs, the palps, which are known to play important roles in gustation. In the locust, recent molecular evidence has shown that nearly all olfactory receptor proteins expressed in the palps are also expressed in the antenna, while electrophysiological studies have shown that olfactory sensory neurons on the palps respond to puffs of diverse categories of volatile chemicals. However, no description exists of neural activity beyond the sensory periphery, and it is not known whether the palp olfactory system uses the same coding principles employed by the antennal olfactory system. We use anatomical and electrophysiological techniques to map and characterize the first three layers of the palp olfactory system of the locust *Schistocerca americana*, starting with palp olfactory sensilla and extending to ensemble activity of third order neurons. We then compare the response properties of palp and antennal olfactory neurons at analogous points along the sensory pathways. We found that the palp system is more narrowly tuned than the antennal system, responding best to food odorants. We also found that the baseline and stimulus-induced firing rates of the 2nd order neurons in the palp system are generally much lower than their antennal counterparts; most palp system neurons we sampled showed reliable subthreshold but not spiking responses to odorants. Finally, we found no evidence that the palp sensory system employs the odor-elicited synchronous oscillatory neural behavior characteristic of the antennal system. We conclude that the palp olfactory system is specialized for food detection and uses coding mechanisms different from those of the antennal olfactory system. Our results provide an opportunity to link the response format of a sensory system to its function.

Disclosures: **Z. Aldworth:** None. **M. Stopfer:** None.

Poster

PSTR144. Olfaction: Peripheral Mechanisms and Behavior

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR144.03/V25

Topic: D.04. The Chemical Senses

Support: DC020519
Karen Toffler Charitable Trust

Title: Heterogeneous concentration-response relationships in the glomerular output of the mouse olfactory bulb

Authors: *D. STORACE[1,2,3], N. SUBRAMANIAN[1];
[1]Biol. Sci., [2]Program in Neurosci., [3]Inst. for Mol. Biophysics, Florida State Univ., Tallahassee, FL

Abstract: Detecting and recognizing relevant odor stimuli requires that an animal be able to recognize an odor as the same across a large range of concentrations, while also maintaining sensitivity to variations in the concentration of the same odor. However, it remains unclear where these perceptual functions are generated in the brain. Glomerular measurements of the olfactory receptor neuron input to the bulb and the apical dendrites of mitral/tufted cells indicate that odor representations are more concentration invariant in the bulb output. Although these results indicate an important role for olfactory bulb processing in generating more stable representations of olfactory stimuli, it remains unclear whether the transformation is heterogeneous across the glomerular population. To test this possibility, we carried out glomerular measurements from the apical dendrites of mitral/tufted cells in response to odors presented across a large concentration range using 2-photon calcium imaging in awake mice. Mitral/tufted glomeruli responded with a heterogeneous mix of concentration-response profiles which were quantified using a monotonicity index. Approximately half of the glomeruli had odor responses that increased monotonically with concentration increases, while the remainder had a range of different nonmonotonic relationships. In contrast, most olfactory receptor neuron concentration-response functions exhibited monotonic relationships with changes in odor concentration. The results support a model in which higher concentrations can activate stronger modulatory processing within the bulb via broad activation across the olfactory receptor neuron glomerular input.

Disclosures: **D. Storace:** None. **N. Subramanian:** None.

Poster

PSTR144. Olfaction: Peripheral Mechanisms and Behavior

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR144.04/W1

Topic: D.04. The Chemical Senses

Support: DFG Grant EG135/6-1, 7-1
DFG Grant FOR 5424

Title: Blood pressure pulsations modulate olfactory bulb activity via mechanosensitive ion channels

Authors: L. JAMMAL SALAMEH[1], S. H. BITZENHOFER[2], I. L. HANGANU-OPATZ[2], M. DUTSCHMANN[3], *V. EGGER[1];
[1]Regensburg Univ., Regensburg, Germany; [2]Univ. Med. Ctr. Hamburg-Eppendorf, Hamburg, Germany; [3]Florey Neurosci. Inst., Melbourne, Australia

Abstract: Intracranial pressure pulsations are known to result from respiration and from the transmission of heartbeat evoked arterial pressure pulsations through the cerebral vascular system. Whether these pulsations can be detected by the central nervous system has been an open question so far. Here, we describe for the first time that arterial pressure pulsations can directly modulate neuronal activity via baroreceptive transduction. We record local field potentials (LFP) within the rat olfactory bulb using a semi-intact, perfused nose brain preparation (NBP), while monitoring the pressure pulsations induced by the peristaltic perfusion pump which happen to fall within the physiological range of heartbeat induced pulsations. Indeed, there are slow LFP oscillations recorded at the mitral cell layer directly correlated to these pulsations (n = 13 NBPs). These oscillations are sensitive to hypoxia (n = 13) and can be dampened by a Windkessel device in the perfusion system (n = 3), but are insensitive to the blockade of neuronal spiking (n = 8). Moreover, we find that mitral cell spiking is in part synchronized to this baroreceptive LFP signal (n = 20). These data let us hypothesize that the heartbeat should exert a modulating influence on OB neuron activity *in vivo*, too. Indeed, parallel recordings of unit activity with a 16-channel multielectrode and heartbeat in awake mice (n = 19 animals) revealed that a subset of OB neurons synchronize their spiking to heartbeat, independently of presence or absence of nasal respiration (n = 6). This modulation is less pronounced and restricted to a smaller fraction of detected units compared to respiratory coupling. Multiple lines of evidence support the notion that cationic fast mechanoreceptors within the mitral cell membrane, most likely Piezo2, play a crucial role in transducing this baroreceptive response: The LFP oscillations were only sensitive to the TRPC/Piezo blocker D-GsMTx4 (n = 13 NBPs, local injection), and not to the broad TRPC channel blocker SKF 96365 (n = 12). Otherwise, GsMTx4 did not reduce spike rate and the overall LFP activity. The observed correlation between changes in LFP oscillatory strength and changes in pressure pulsation frequency (n = 20 NBPs) is in line with differential mechanosensing, and the waveform of the slow LFP oscillations derived from the observed spectral harmonics (n = 53 NBPs) are best explained by Piezo2 gating properties, rather than those of Piezo1. We propose that this intrinsic interoceptive mechanism within the brain can directly modulate olfactory sensitivity e.g. during arousal.

Disclosures: L. Jammal Salameh: None. **S.H. Bitzenhofer:** None. **I.L. Hanganu-Opatz:** None. **M. Dutschmann:** None. **V. Egger:** None.

Poster

PSTR144. Olfaction: Peripheral Mechanisms and Behavior

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR144.05/W2

Topic: D.04. The Chemical Senses

Support: Francis Crick Institute Grant FC001153
UK Medical Research Council Grant MC_UP_1202/5
Wellcome Trust Investigator Grant 110174/Z/15/Z
Physics of Life Grant EP/W024292/1
European Research Council, European Union's Horizon 2020 Research and Innovation Programme grant 852455
Swiss Light Source, beamline TOMCAT proposal 20190417
Swiss Light Source, beamline TOMCAT proposal 20211896
Swiss Light Source, beamline TOMCAT proposal 20220191
European Synchrotron Radiation Facility, beamline ID16A proposal ls2918
European Synchrotron Radiation Facility, beamline ID16A proposal ls3025
European Synchrotron Radiation Facility, beamline ID16A proposal ls3186

Title: Odour responses of sister projection neurons in the mouse olfactory bulb, revealed by correlative two-photon imaging and synchrotron X-ray holographic nano-tomography

Authors: *Y. ZHANG[1,2], C. BOSCH[1], T. ACKELS[1,2], A. BONNIN[3], A. NATHANSEN[4], C. WALTENBERG[5], N. RZEPKA[4], M. BERNING[4], A. LAUGROS[6], J. LIVINGSTONE[6], P. CLOETENS[6], A. PACUREANU[1,2,6], A. T. SCHAEFER[1,2];
[1]Sensory Circuits and Neurotechnology Lab., The Francis Crick Inst., London, United Kingdom; [2]Dept. of Neuroscience, Physiol. and Pharmacol., Univ. Col. London, London, United Kingdom; [3]Paul Scherrer Inst., Villigen, Switzerland; [4]scalable minds GmbH, Potsdam, Germany; [5]Carl Zeiss Microscopy GmbH, Oberkochen, Germany; [6]ESRF, The European Synchrotron, Grenoble, France

Abstract: In mammals, the olfactory bulb (OB) conducts the first layer of olfactory information processing. Axons from olfactory sensory neurons from the nose aggregate by olfactory receptor type into neuropil structures called glomeruli, where they synapse onto apical dendrites of OB projection neurons (PN). Each PN sends its apical dendrite to only one glomerulus, sharing excitatory drive with several sister PNs and outputs to other brain areas.
Glomerulus size and the number of sister PNs vary, however, it is unclear how sister PNs respond to odours, and what computational benefit this brings. Understanding the responses of sister PNs to controlled odour stimuli will provide mechanistic insights into how the OB transforms olfactory information, and into the functional stereotypy of mammalian cell types.

We recorded responses of 100s of PNs expressing GCaMP6f to a panel of 50 monomolecular odorants with volumetric *in vivo* two-photon (2P) imaging in anaesthetized mice. We then chemically fixed and heavy metal stained the tissues [1, 2]. The tissues, ~3x3x0.5 mm in size, contain the 2P imaged volume and were subsequently imaged using synchrotron X-ray computed tomography with propagation-based phase contrast (SXRT, n = 18) [1, 2]. The SXRT datasets allowed us to precisely locate previously imaged cells and trim the sample to ~1x1x0.5 mm for synchrotron X-ray holographic nano-tomography (XNH, n = 3) [3, 4]. For inter-animal comparisons, all n = 3 samples with complete *in vivo* 2P, SXRT and XNH datasets were functionally imaged around a genetically targeted glomerulus (MOR174/9) [5]. >80% of all functionally imaged cells were found in the SXRT and XNH datasets and their apical dendrites traced back to their parent glomeruli.
For each animal, we characterised the odour responses of ~20 glomeruli, each containing 1-7 sister PNs. Odour responses of sister PNs were highly correlated, especially for strongly activating odours. The correlation dropped as the strongest odours were omitted. Moreover, the odour response profile of PNs predicted their parent glomerulus identity reliably. This function-structure approach allowed, for the first time, an unbiased mapping of the parent glomeruli of PNs using structural ground-truth. It reveals a high degree of similarity of sister cell PNs' responses to monomolecular odorants, and showcases the power of a combined physiology and structure approach to understand mammalian neural circuits.

1. C. Bosch *et al.*, *Nat. Commun.* 13, 2923 (2022).
2. Y. Zhang *et al.*, *Front. Cell Dev. Biol.* 10, 880696 (2022).
3. C. Bosch *et al.*, *Appl. Phys. Lett.* 122, 143701 (2023).
4. A. T. Kuan *et al.*, *Nat Neurosci.* 23, 1637-1643 (2020).
5. D. Schwarz *et al.*, *Nat Commun.* 9, 183 (2018).

Disclosures: **Y. Zhang:** None. **C. Bosch:** None. **T. Ackels:** None. **A. Bonnin:** None. **A. Nathansen:** None. **C. Waltenberg:** None. **N. Rzepka:** None. **M. Berning:** None. **A. Laugros:** None. **J. Livingstone:** None. **P. Cloetens:** None. **A. Pacureanu:** None. **A.T. Schaefer:** None.

Poster

PSTR144. Olfaction: Peripheral Mechanisms and Behavior

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR144.06/W3

Topic: D.04. The Chemical Senses

Support: NSF IOS Grant 2114775
NIH Grant DC-016293

Title: Serotonin targets discrete inhibitory motifs within the antennal lobe of Drosophila

Authors: *A. M. DACKS[1], J. D. RALSTON[1], F. SALMAN[1], O. M. COOK[1], T. R. SIZEMORE[2], J. JONAITIS[1];

[1]Biol., West Virginia Univ., Morgantown, WV; [2]Dept. of Molecular, Cell. and Developmental Biol., Yale Univ., New Haven, CT

Abstract: Neuromodulation allows the nervous system to flexibly process sensory information by differentially adjusting the function of individual network components. Newly available large EM datasets have allowed for the quantification of connectivity of individual modulatory neurons, revealing key targets for neuromodulation within a network. However, without knowing the modulatory receptors expressed by individual cellular components, the impact of neuromodulation on network activity is difficult to predict. We recently completed a connectomic analysis which revealed that, within the antennal lobe ("AL") of Drosophila, serotonergic modulatory neurons predominantly target only ~25 highly interconnected local interneurons (LNs) from three morphological LN types. We sought to determine if these three LN types represented discrete functional network components as well as the receptors expressed by each LN type. While all were GABAergic, one LN type co-expressed neuropeptides, and the three LN types differed in the principal AL demographics upon which they synapse, as well as the spatiotemporal nature of their odor response properties. Furthermore, we determined that each LN type also differed in the serotonin, GABA and neuropeptide receptors that they each express. Collectively, this combined connectivity and receptor expression analysis suggests that serotonergic neurons in Drosophila differentially modulate distinct inhibitory motifs of an interconnected LN network within the olfactory system, and provides an expansion from connectomic data to receptor expression and functional responses.

Disclosures: A.M. Dacks: None. **J.D. Ralston:** None. **F. Salman:** None. **O.M. Cook:** None. **T.R. Sizemore:** None. **J. Jonaitis:** None.

Poster

PSTR144. Olfaction: Peripheral Mechanisms and Behavior

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR144.07/W4

Topic: D.04. The Chemical Senses

Support: NIH DC019124

Title: Modulation of olfactory behavior and neural responses by 5HT

Authors: *O. ESCANILLA[1], M. EINHORN[2], D. SCOTT[3], M. LEWIS[3], C. LINSTER[1]; [2]Dept. Psychology, [3]Neurobio. and Behavior, [1]Cornell Univ., Ithaca, NY

Abstract: Odor processing in the olfactory bulb is modulated by centrifugal inputs from cortical networks as well as neuromodulatory nuclei. While cholinergic and noradrenergic modulation of olfactory bulb processing has been well studied, few studies have investigated modulation by 5HT. We used a multi-experiment approach to investigate the function of 5HT in the olfactory bulb (OB) for odor detection and odor discrimination: four behavioral experiments, recordings in awake behaving rats, and in mouse olfactory bulb slices. Behavioral experiments show that

additional 5HT in the OB enhances odor detection and discrimination at very low concentrations, blockade of 5HT receptors increases odor thresholds significantly. Overall concentration invariance is improved by boosting responses to low concentrations, rather than decreasing responses to high concentrations as proposed by through other mechanisms. In vivo and brain slice recordings show an overall increase in olfactory bulb spontaneous and odor evoked responses accompanied by an increase in OB dynamics. These effects translate to better read out and more significant odor responses in olfactory cortex of awake behaving rats, limited to very low concentration odorants. Computational modeling of known cellular and synaptic effects show that at low odor concentrations increased neural firing, restricted to specific phases of the gamma oscillation cycle increase cortical responses to low concentration odorants. These results are in agreement with recordings in anterior piriform cortex showing a higher number of significant odor responses when 5HT is infused into the OB. Overall our results show that modulation of neural activity in the OB has strong effects on cortical responses which reflect behavioral observations. 5HT modulation helps concentration invariance by boosting responses to low concentration stimuli.

Disclosures: O. Escanilla: None. **M. Einhorn:** None. **D. Scott:** None. **M. Lewis:** None. **C. Linster:** None.

Poster

PSTR144. Olfaction: Peripheral Mechanisms and Behavior

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR144.08/W5

Topic: D.04. The Chemical Senses

Support: IOS-1656830 NSF

Title: Axially decoupled optical control and readout of activity for mapping functional connectivity of neural circuits

Authors: *D. ALBEANU[1], M. KOH[1], F. ANSELMI[1], S. TEJA[1], W. G. BAST[2], H. CHAE[1], P. GUPTA[1], D. HERNANDEZ TREJO[1], P. VILLAR[1], M. B. DAVIS[1], A. BANERJEE[1]; [2]Neurosci., [1]Cold Spring Harbor Lab., Cold Spring Harbor, NY

Abstract: Coupling multiphoton imaging and optogenetic patterned photo-stimulation strategies *in vivo* provides a conduit for investigating the function of neural circuits in 3D. We developed a high-throughput, cost-effective strategy for flexible, axially-decoupled optical control and readout of neural activity (ADCOR). By optically conjugating a digital micro-mirror device (DMD) illuminated by a coherent light source with a motorized holographic diffuser, we achieved axial optical sectioning (*30 µm z-resolution, across 1.20 x 1.50 mm*) of wide field patterned photo-stimulation. As proof of principle, we controlled the activity of individual input glomeruli on the olfactory bulb surface, while monitoring the responses of mitral and tufted output neurons within the same plane (dendritic tufts) and down to 250 µm below brain surface.

Photo-stimulation of olfactory sensory neuron presynaptic terminals selectively evoked responses in the targeted glomeruli, enabled the identification of sister mitral and tufted somata, and also revealed sparse lateral inhibitory interactions. In addition, optogenetic activation of glomerular gabaergic/dopaminergic (DAT+) interneurons during odor sampling triggered heterogeneous suppression of mitral cell responses, suggesting specificity in the inhibitory connectivity of bulbar circuits. In summary, we report a high-throughput all-optical physiology method readily applicable for interrogating layer-organized neural circuits.

Disclosures: D. Albeanu: None. **M. Koh:** None. **F. Anselmi:** None. **S. Teja:** None. **W.G. Bast:** None. **H. Chae:** None. **P. Gupta:** None. **D. Hernandez Trejo:** None. **P. Villar:** None. **M.B. Davis:** None. **A. Banerjee:** None.

Poster

PSTR144. Olfaction: Peripheral Mechanisms and Behavior

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR144.09/W6

Topic: D.04. The Chemical Senses

Support: Western Michigan University

Title: Neuroplasticity in the zebrafish olfactory bulb after immune modulation

Authors: *B. EBENDICK[1], S. VAR[2], C. BYRD-JACOBS[1];
[1]Biol., Western Michigan Univ., Kalamazoo, MI; [2]Univ. of Minnesota, MINNEAPOLIS, MN

Abstract: Recovery from neuronal injury includes clearance of damaged tissues and cell regeneration, but not all animals exhibit equal neuroplasticity. For many animals it is a slow or incomplete process, but zebrafish demonstrate rapid and persistent recovery. The primary immune cells of the brain, phagocytic microglia, are active during both pro- and anti-inflammatory functions, but their role in zebrafish neuroplasticity is unclear. Modulating microglial populations either to decrease or increase their activity illustrates the contribution of these cells on the timing of recovery. L-clodronate targets phagocytic cells for apoptosis, and zymosan, a sterile yeast extract, stimulates immune cells. In addition, the timing of injection of immune-modulating compounds may affect microglial activity in recovery. Our lab has shown that chemical lesions of the olfactory epithelium in zebrafish cause significant tissue damage that resolves by 7 days. Pre-treatment groups received L-clodronate, zymosan, or saline injections starting 24 hours prior to lesioning. Concurrent treatment groups received injections immediately prior to lesioning. Baseline fish were lesioned without injection. Detergent was used to lesion sensory neurons innervating the right olfactory bulb, preserving the left side as an internal control. Three glomerular structures in the bulb were visualized with antibody-labeled sensory axons in whole brains using confocal microscopy. Structures were assessed on a 4-point scale for damage from 1 to 7 days post-lesioning (dpl) and compared to control groups. Saline-treated fish responded similarly to untreated controls: damage peaked early and structures were reorganized

by 7 dpl. Drug pretreatments, however, significantly affected the recovery timeline. A delayed recovery after clodronate was expected; however, clodronate-pretreated fish appeared recovered in all three glomerular structures by 4 dpl, significantly faster than baseline. Zymosan pretreatment showed very little damage at 1 dpl, peaked at 4 dpl (later than other groups), and returned to control morphology by 7 dpl. In both pretreatments, the timeline was contracted or shifted from baseline. This suggests a role for microglia in the timing of inflammatory response. The activity of microglia after neuronal damage can vary dramatically during recovery, from persistent inflammation to resolution. Zebrafish as a model system can demonstrate how conserved immune system features promote recovery in the nervous system of adult mammals, leading to potential treatments for brain injury and disease.

Disclosures: **B. Ebendick:** None. **S. Var:** None. **C. Byrd-Jacobs:** None.

Poster

PSTR144. Olfaction: Peripheral Mechanisms and Behavior

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR144.10/W7

Topic: D.04. The Chemical Senses

Support: Grant-in-Aid for Scientific Research (C) 22K06490
OIST Graduate University

Title: Probing stimulus encoding in the olfactory bulb using synthetic olfactory stimuli

Authors: *X. FU, J. K. REINERT, S. LINDEMAN, I. FUKUNAGA;
Okinawa Inst. of Sci. and Technol. Grad. Univ., Okinawa, Japan

Abstract: An essential task for the sensory systems of the brain is to encode external stimuli as patterns of neuronal activity. In the mouse, complex synaptic processing of odor information starts in the olfactory bulb. The output of this region is carried by at least two classes of neurons, mitral and tufted cells (M/TCs). These neurons are thought to differentially encode odors, the former using spike timing and the latter using rate coding. However, a direct, causal demonstration of this is currently lacking.
We aimed to explore how M/TC activation patterns relate to olfactory perception using sniff-locked optogenetic stimuli in head-fixed mice expressing ChR2 in M/TCs (Tbet-Cre::Ai32 mice) performing a Go/No-Go task. We built a system for temporally precise optogenetic stimulation, where light presentations are triggered based on online analysis of sniff signals. The temporal precision of the stimuli in behaving mice, as measured by the standard deviation in the light onset from the start of inhalation, was 5.9 ms (approximately 2.4% of an average sniff interval; n = 511 light trials from 10 mice). We used a single optical fiber targeting the anterior piriform cortex to activate the M/TC axons, to reliably evoke action potential in both neuron ytpe at all sniff phases.
Using this system, we aimed to investigate how distinct synthetic olfactory stimuli are perceived.

Our expectation is that mice can distinguish different numbers of light pulses, as well as phase differences, when MCs and TCs are both activated. We first trained the mice on a simple olfactory discrimination task, so that they acquire the Go/No-Go rule. Once proficient, they were trained to discriminate between rewarded and unrewarded optogenetic stimuli. We first demonstrate that the mice can distinguish between trials with and without optogenetic stimuli (trials to d' = 2 for detecting 5 pulses = 111 ± 39 trials, and to detect 1 pulse of light = 96 ± 68 trials; n = 4 mice), and second, demonstrate that they distinguish between different numbers of light pulses (trials to criterion for 3 pulses vs. 1 pulse = 110 trials, n = 1 mouse). Curiously, the mice failed to distinguish between stimuli presented during the inhalation vs. exhalation phases (d' after 180 trials of training = 0.13 ± 0.10; n = 3 mice). This failure to distinguish the phase-specific stimuli may occur because M/TCs are stimulated simultaneously here. That is, the timing-based code may work only in the presence of a separate, reference signal. We are currently testing the hypothesis that the tufted cells carry this reference signal. We aim to ultimately reveal how olfactory stimuli are encoded by the parallel olfactory pathways.

Disclosures: **X. Fu:** None. **J.K. Reinert:** None. **S. Lindeman:** None. **I. Fukunaga:** None.

Poster

PSTR144. Olfaction: Peripheral Mechanisms and Behavior

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR144.11/W8

Topic: D.04. The Chemical Senses

Support: NSF GRFP
Brain and Behavior Research Foundation
Zuckerman Institute Recruitment Fund
Whitehall Foundation Grant
Hypothesis Fund Grant
Columbia University RISE Grant
EE Just Grant
Columbia University Provost's Award

Title: Determining the molecular mechanisms of heritable stress-induced bias in olfactory neuron differentiation

Authors: *R. STECKY, S. LOMVARDAS, B. MARLIN;
Columbia Univ., New York, NY

Abstract: Human history gives us myriad examples of intergenerational inheritance of traumatic experience (Heijmans et al., 2008; Yehuda et al., 2016). Recently, these population-wide examples have been supported with experimental data that suggest stress-induced somatic changes can be inherited across generations (Dias and Ressler, 2014; Shea et al., 2015). Using the mouse model, the Marlin lab has found that pairing an odor with a mild foot shock (a

behavioral paradigm called olfactory fear conditioning) induces an upregulation in the birth of olfactory sensory neurons that respond to the paired odor. This phenotype has been demonstrated in both conditioned mice and their unconditioned progeny (Liff et al., 2023). How does odor-shock pairing bias the otherwise-stochastic differentiation of newly born neurons? I aim to identify molecular mechanisms of this observed upregulation phenotype using transcriptomics. One day after conditioning with the P2 ligand, I performed bulk RNA-sequencing on isolated P2+ mature neurons in order to identify differentially expressed genes between odor-shock paired mice and the unpaired control. Using a generalized linear model to compare the two groups (DESeq2, Love et al., 2014), I found several genes of the *Bpif* family to be upregulated in the paired P2+ neurons (n = 4, mean log2fc = 5.879, mean p-value = 0.017). This result suggests a capacity for coincidence detection in these neurons. That is, they appear to possess the ability to detect the simultaneity of the P2 ligand and the foot shock and to alter their gene expression in response. I hypothesize that the aforementioned differentially expressed genes function as part of an intercellular communication mechanism that biases neighboring stem cells to develop into P2+ neurons. To follow, I performed bulk RNA-sequencing on olfactory stem cells seven days after conditioning with the P2 ligand. Despite lacking the molecular maturity to detect the P2 ligand, these cells display broad differential gene expression between the paired group and the unpaired control (n = 4, 1216 individual genes, mean log2fc = -0.625 ± 0.029 SEM, mean adjusted p-value = 0.014). I hypothesize that this effect is a cell-nonautonomous response to the biasing message from paired P2+ neurons. In sum, these data suggest an intercellular mechanism of communication between P2+ mature neurons and the undifferentiated stem cell population, which functions to bias the neuronal differentiation process. These findings have profound implications in helping us understand how the olfactory system integrates salient environmental information and how traumatic experiences can regulate gene expression in a heritable manner.

Disclosures: **R. Stecky:** None. **S. Lomvardas:** None. **B. Marlin:** None.

Poster

PSTR144. Olfaction: Peripheral Mechanisms and Behavior

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR144.12/W9

Topic: D.04. The Chemical Senses

Support: Brain and Behavior Research Foundation Grant 30380
Zuckerman Institute Recruitment Fund
Whitehall Foundation Grant
Hypothesis Fund Grant
Columbia University RISE Grant
EE Just Grant
Columbia University Provost's Award

Title: Olfactory fear conditioning biases olfactory stem cell receptor fate

Authors: *C. W. LIFF, Y. R. AYMAN, E. C. B. JAEGER, H. S. LEE, A. KIM, B. J. MARLIN;
Zuckerman Inst., Columbia Univ., New York, NY

Abstract: The main olfactory epithelium initiates the process of odor encoding. Recent studies have demonstrated intergenerationally inherited changes in the olfactory system in response to fear conditioning, resulting in increases in olfactory receptor frequencies and altered responses to odors. We investigated changes in the morphology of the olfactory sensory epithelium in response to an aversive foot stimulus. Here, using two distinct ligand-receptor pairs, we achieve volumetric cellular resolution to demonstrate that olfactory fear conditioning increases the number of odor-encoding neurons in mice that experience shock-odor conditioning (F0; n=11,12; $p<0.001$), as well as their naïve offspring (F1; n=12,14; $p<0.001$). Using EdU-labeling to measure birth rates of olfactory sensory neurons, we provide evidence that biased stem cell receptor choice contributes to these increases in F0 (n=6,6; $p<0.001$). Interestingly, we do not observe the inheritance of active avoidance of the conditioned odor, contrary to prior studies demonstrating inherited behavior changes. Thus, we reveal dynamic regulation of the main olfactory epithelium receptor composition in response to olfactory fear conditioning, providing insight into the heritability of acquired phenotypes.

Disclosures: C.W. Liff: None. **Y.R. Ayman:** None. **E.C.B. Jaeger:** None. **H.S. Lee:** None. **A. Kim:** None. **B.J. Marlin:** None.

Poster

PSTR144. Olfaction: Peripheral Mechanisms and Behavior

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR144.13/W10

Topic: D.04. The Chemical Senses

Title: Combinatorial allosteric interactions at odorant receptors for odor mixture coding

Authors: *K. FUKATA, S. INAGAKI, B. SAHA, T. IMAI;
Kyushu Univ., Fukuoka, Japan

Abstract: Sensory signals from the environment are mostly comprised of multiple types of stimuli. For example, natural odors are mostly composed of various kinds of odorants. The binding of an odorant to odorant receptors (ORs) activates or inhibits olfactory sensory neurons (OSNs) and generates the combinatorial receptor code at the olfactory epithelium. However, OSN responses to odor mixtures are not necessarily the sum of the responses to their components. OSN responses to an odorant are often modulated by another odorant, known as antagonism and synergy. Previous studies have revealed the mechanism of antagonism, which occurs at the level of the ORs. However, the mechanisms underlying synergistic responses have remained unclear.
Here, we examined the hypothesis that an odorant acts as an allosteric modulator for ORs. We evaluated the responses of OSNs to odorants using in vivo two-photon calcium imaging of the

olfactory epithelium. We first examined the concentration-response curves for amyl acetate and pentanal in each of the OSNs. We then examined how the concentration-response curves to an agonist were modulated by a non-agonist. We observed synergistic effects in 21.4% of the agonist-responsive OSNs. The response curves showed leftward or upward shifts with the addition of the non-agonist, suggesting allosteric enhancement at the ORs.
We next investigated the ligand specificity at the allosteric sites using three aliphatic aldehydes (butanal, pentanal, and hexanal). An amyl acetate responses were allosterically enhanced by unique and specific sets of aldehydes in each OSN: it was broadly tuned in some OSNs but narrowly tuned in others, suggesting OR-specific tuning of the allosteric binding sites. Previously, only agonist binding sites (orthosteric sites) were considered for ORs. Accordingly, the "combinatorial receptor code" has been proposed based only on the odorant binding to the orthosteric sites. However, our study suggests that many ORs also have allosteric sites with different ligand specificities, as is known for some G protein-coupled receptors. Thus, the combinatorial receptor code for an agonist can be further modulated by the combinatorial binding of allosteric modulators. Allosteric modulation adds another layer of combinatorial coding to produce diverse responses to odor mixtures.

Disclosures: **K. Fukata:** None. **S. Inagaki:** None. **B. Saha:** None. **T. Imai:** None.

Poster

PSTR144. Olfaction: Peripheral Mechanisms and Behavior

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR144.14/W11

Topic: D.04. The Chemical Senses

Support: U19 NS107464-01

Title: Behaviorally relevant features of the neural code in olfactory bulb

Authors: *S. KARIMIMEHR[1,3], S. CEBALLO CARPENTIER[3], M. KARADAS[3], D. RINBERG[3], *S. KARIMIMEHR[1], *S. KARIMIMEHR[2];
[1]New York Univ., New York, NY; [2]New York Univ., Woodside, NY; [3]Neurosci. Inst., NYU Langone Hlth., New York, NY

Abstract: Odor stimuli evoke spatiotemporal patterns of activity at multiple levels in the olfactory bulb, the Glomeruli and subsequent mitral/tufted (MT) cells. How the similarities between patterns of neural activity are related to perceptual similarities between sensory stimuli is still debatable. In this work, we designed an experiment using the 2-alternative-forced-choice paradigm (2AFC) to measure the generalization ability of mice during precise odor discriminations. In this task, we are able to smoothly vary spatiotemporal patterns of activity using three component odor mixtures to identify the relevant features of the neural activity that drive behavioral discriminations.
During this task, we trained mice to discriminate a specific mixture of three odors (referred to as

the "Target") from a range of different odor stimuli (referred to as "non-Targets"). In subsequent probe trials, we manipulated the composition of the mixture to test the mice's ability to generalize their response. Then, we used two photon Ca2+ imaging and measured the neural activity in mice expressing a fast calcium indicator (GCaMP6f) in post synaptic glomeruli and MT cells. Based on these recordings, we will identify the crucial aspects of neural activity that drive behavior. Our preliminary data showed a strong correlation between the temporal order of glomerular/MT activation in probe trials and behavioral performance. Specifically, we discovered that this correlation exists within a window of less than 200ms. Glomeruli that were activated beyond this window may not contribute to the behavioral correlation with neural activity.
These findings provide valuable insights into how the olfactory system represents and distinguishes between odor mixtures. The importance of the order of neural activity emphasizes the significance of temporal dynamics in encoding odor mixtures.

Disclosures: S. Karimimehr: None. **S. Ceballo Carpentier:** None. **M. Karadas:** None. **D. Rinberg:** None. **S. Karimimehr:** None. **S. Karimimehr:** None.

Poster

PSTR144. Olfaction: Peripheral Mechanisms and Behavior

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR144.15/W12

Topic: D.04. The Chemical Senses

Support: NIH BRAIN Initiative Grant U19NS112953
Simons Foundation, Simons Society of Fellows Junior Fellow, Grant 965381

Title: Revealing the neural signature of perceived odor intensity.

Authors: *B. BARRA[1,2], J. HARVEY[2,1], T. REIZIS[2,1], J. MAINLAND[3], D. RINBERG[2,4]; [1]New York Univ. Sch. of Med., New York, NY; [2]Neurosci. Inst., NYU Langone Hlth., New York, NY; [3]Monell Chem. Senses, Philadelphia, PA; [4]Ctr. for Neural Sci., New York Univ., New York, NY

Abstract: How the brain represents physical properties of sensory stimuli is a central question in sensory systems neuroscience. In olfaction, the concentration of odorant molecules in the air translates to perceived odor intensity. Currently, how perceived intensity is encoded in the olfactory system is poorly understood. Studies have shown that changes in odorant concentration are correlated with changes in firing rate, temporal shifts relative to inhalation, overall synchrony of neural responses, and the activation of different ensembles of responsive neurons. However, odorant concentrations do not unambiguously correspond to perceived intensity: at similar concentrations some odors evoke very strong sensations, while others are barely perceivable. Therefore, it remains unclear which neural features underlie the perception of odor intensity. One

major challenge in the study of the neural encoding of intensity consists in obtaining concurrent perceptual reports and neural recordings from animal models. Here, we approached this problem by adopting an existing behavioral paradigm developed in rats[1] with the aim to find which odorant concentrations produced percepts that matched in intensity in mice. Mice were trained to discriminate 8 concentrations of an odor within the same order of magnitude. After training, mice could discriminate the four lowest and the four highest concentrations, by using a trained decision boundary. Interestingly, when a second odor was introduced in the paradigm, animals showed odor-specific changes of behavior that resulted in an odor-specific shift of decision boundary. We interpret this shift to reflect a difference in perceived intensity between the two odors. Conversely, overlapping decision boundaries would indicate that the odorants at those concentrations were perceived as equally intense. We used this paradigm to estimate intensity matched concentrations of n=3 odors, at n=3 different concentration ranges. Then, we used wide field calcium imaging to record glomerular activity in the olfactory bulb. For each odor, we recorded spatio-temporal patterns of glomerular activity during 2s-long odor presentations, across a vast range of molar concentrations (from 10^{-7} to 10^{-1}). We then combined neural recordings and behavioral results to investigate which neural features are predictive of perceived odor intensity, i.e. are correlated to concentration changes, while holding stable across intensity-matched odors. We used this integrated approach to gain insights about the neural encoding of odor intensity. (1) Wojcik, P. T. & Sirotin, Y. B. Single Scale for Odor Intensity in Rat Olfaction. *Curr. Biol.* **24**, 568-573 (2014)

Disclosures: **B. Barra:** None. **J. Harvey:** None. **T. Reizis:** None. **J. Mainland:** F. Consulting Fees (e.g., advisory boards); Osmo Labs, PBC. **D. Rinberg:** Other; Founder and a chief scientific adviser of the company Canaery, Inc.

Poster

PSTR144. Olfaction: Peripheral Mechanisms and Behavior

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR144.16/W13

Topic: D.04. The Chemical Senses

Support: U19NS107464
U19NS112953

Title: Functional interactions between olfactory circuits shapes the timing and structure of odor representations

Authors: *M. KARADAS[1], J. GILL[1], S. CEBALLO[1], S. SHOHAM[2,1], D. RINBERG[3,1]; [1]NYU Langone Hlth., New York, NY; [2]Inst. for Engin. in Biomedicine, New York Univ. Sch. of Med., New York, NY; [3]New York Univ., New York, NY

Abstract: Odors evoke distinct patterns of neural activity across different concentrations, both at the input level of olfactory sensory neurons (OSN) and glomeruli, as well as the next level,

mitral/tufted cells (MTCs). What features of these patterns carry invariant information about odor identity and what are the neural mechanisms defining these features? To address this question, we designed an all-optical system to monitor the activity of many glomeruli and MT cells with high temporal resolution and developed an optogenetic method for establishing functional connectivity between glomeruli and MTCs. We determined the transformation of odor responses from glomeruli to their MTCs. We combined two photon Ca2+ imaging with one photon patterned optogenetics in mice expressing a fast calcium indicator (GCaMP6f) in glomeruli and MTCs, and a light-sensitive opsin (ChR2) in OSNs. We found that MTCs connected to early activated glomeruli exhibited stereotypic excitatory responses following their parent glomeruli. At the same time, MTCs of later activated glomeruli mostly have inhibitory responses, although their glomeruli exhibit a well-pronounced excitatory response to the odors. Then, we determined the responsiveness of MTCs to their glomerular activation by employing brief optogenetic pulses in the presence of odor stimuli. Our finding revealed that MTCs can effectively transmit glomerular signals to the cortex efficiently, relaying glomerular signals to the cortex, but only within a limited temporal range at the onset of the sniff cycle. These findings reveal potential mechanisms for concentration invariant odor coding, and support the primacy model for olfactory information processing, wherein the earlier-activated glomeruli carry odor identity information. Additionally, we used odor and light stimuli targeting individual glomeruli to assess the degree of stereotypy and diversity among its sister MTC. In our experiment, we observed the robust stereotypic response pattern among sister mitral cells for comprehensive set of stimuli. Using patterned optogenetic of glomeruli sequences, we further investigated the distribution of the inhibitory connections across multiple glomeruli, aiming to establish the specificity of these connections.
Together, our approach and finding will provide essential insight into the principles and mechanisms of odor processing in the olfactory bulb.

Disclosures: **M. Karadas:** None. **J. Gill:** None. **S. Ceballo:** None. **S. Shoham:** None. **D. Rinberg:** Other; Founder and a chief scientific adviser of the company Canaery, Inc.

Poster

PSTR144. Olfaction: Peripheral Mechanisms and Behavior

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR144.17/W14

Topic: D.04. The Chemical Senses

Support: NIH Grant U19 NS112953

Title: Mice share a common manifold for representing odor quality

Authors: *J. S. HARVEY[1], K. SAMOILOVA[2], H. NAKAYAMA[1], F. PASHAKHANLOO[2], A. KOULAKOV[2], D. RINBERG[1];
[1]Neurosci. Inst., NYU Sch. of Med., New York, NY; [2]Cold Spring Harbor Lab., Cold Spring Harbor, NY

Abstract: Understanding how odor space is constructed and shaped by the olfactory system will allow us to decode olfactory information from neural recordings, and align representations across multiple animals. Here, we compare odor quality spaces constructed from both neural recordings and perceptual judgments obtained in mice. We use wide-field calcium imaging to monitor olfactory sensory neurons in the mouse olfactory bulb, obtaining spatio-temporal patterns of glomerular activity in response to 24 odors. We find Kendall-tau rank correlation of glomerular sequence provides a distance metric by which a concentration-invariant odor quality space can be constructed, allowing cross-concentration odor decoding in only 6 or 7 dimensions.
We then use this metric to align odor representations across animals. We find that non-linear kernel manifold alignment requires fewer embedding dimensions than linear methods (PCA and Procrustes), when cross-animal odor prediction is evaluated with leave-one-out cross-validation (LOOCV). We explore cross-animal alignment using a subset of anchor odors, and develop a heuristic that reliably predicts which subsets of odors will provide the best alignment performance. With optimal anchors, just 6 labeled odors are required to align and decode representations of 24 odors across 7 mice with more than 80% accuracy.
Finally, we explored the relationship between neural responses and perceptual similarity, measuring perceptual distances in a cohort of 10 mice using a delayed match-to-sample (DMTS) behavioral paradigm. We find that perceptual spaces can be aligned across individual mice, suggesting that odor relations are perceived similarly by different mice. We show that this common perceptual space can be aligned to the common neural space, allowing odor prediction performance above 80% in 7 dimensions when evaluated with LOOCV.
Overall, we show that odor quality is represented in mice by a common low-dimensional manifold, both on the level of neural responses and odor percepts, suggesting that odor similarity relationships are preserved from sensory transduction through perception.

Disclosures: J.S. Harvey: None. **K. Samoilova:** None. **H. Nakayama:** None. **F. Pashakhanloo:** None. **A. Koulakov:** None. **D. Rinberg:** Other; founder and a chief scientific adviser of the company Canaery, Inc.

Poster

PSTR144. Olfaction: Peripheral Mechanisms and Behavior

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR144.18/W15

Topic: D.04. The Chemical Senses

Support: U19NS112953
U19NS107464
Leon Levy Scholarship in Neuroscience

Title: The geometry and role of sequential activity in olfactory processing.

Authors: *J. GILL[1], M. KARADAS[1], S. SHOHAM[3,2], D. RINBERG[4,1,5];
[1]Neurosci. Inst., [2]Tech4Health Inst., NYU Langone Hlth., New York, NY; [3]Dept. of

Opthalmology, New York Univ. Sch. of Med., New York, NY; [4]Ctr. for Neural Sci., [5]Physics, New York Univ., New York, NY

Abstract: Animals depend on their senses for survival. Mice, who rely on olfaction to navigate the world, can rapidly identify odors within a single sniff across a wide range of concentrations. In the mouse olfactory bulb (OB), mitral and tufted cells (MTCs) respond to odors by changing both the rate and timing of spikes relative to inhalation, resulting in reliable, odor specific sequences that evolve over a single sniff. However, it remains unknown how sequential MTC activity is organized. Specifically, what defines the order in which MTCs fire, and what information is encoded in these sequences? To address this, we performed 2-photon (2P) calcium imaging of hundreds of MTCs expressing the fast calcium indicator jGCaMP8f, permitting us to monitor the sub-sniff timing of responses to a diverse battery of odors. We constructed a space of MTC tuning using the pairwise correlations between MTC odor responses averaged over a single sniff. We then analyzed the propagation of sequences in this space and discovered that sequences originated in a set of similarly tuned neurons and propagated to more distantly tuned neurons, so that the latency of MTC activation was linearly related to distance in tuning space. Analyzing the concentration dependence of sequence propagation, we found that the early part of the sequences carried information that was concentration invariant, while later MTC responses were inconsistent across concentrations. Finally, inspired by the discovery that similarly tuned MTCs are activated sequentially across odors, we constructed and analyzed a computational model for sequence-based unsupervised training of synapses from MTCs to the piriform cortex, which revealed that sequential activity across the entire sniff permits perceptual generalization for novel odors.

Disclosures: J. Gill: None. **M. Karadas:** None. **S. Shoham:** None. **D. Rinberg:** Other; Founder and a chief scientific adviser of the company Canaery, Inc.

Poster

PSTR144. Olfaction: Peripheral Mechanisms and Behavior

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR144.19/W16

Topic: D.04. The Chemical Senses

Support: Stowers Institute for Medical Research
NIH R01 DC0080003
NIH R01 DC020368

Title: Dissecting the neural circuitry underlying processing of mouse female pheromones

Authors: *T. T. CHIREMBA[1], L. MA[1], H. WILSON[1], C. MCKINNEY[1], Z. YU[1], C. R. YU[1,2]; [1]Stowers Inst. for Med. Res., Kansas City, MO; [2]Dept. of Anat. and Cell Biol., Univ. of Kansas Med. Ctr., Kansas City, KS

Abstract: The rodent vomeronasal system mediates pheromone signaling to induce stereotyped social behaviors and neuroendocrine responses. Two large gene families, vomeronasal type 1 receptor (V1r) and vomeronasal type 2 receptor (V2r), encode more than 300 putative pheromone receptor genes in the mouse vomeronasal organ. Despite decades of studies, the specific roles of individual receptors and the neural circuits transmitting pheromone signals have remained largely unknown. Previously, we have identified two members of the V1re clade receptors (V1re9 and V1re12) that detect mouse female urine specific cues, and two members of the V1rj clade (V1rj2 and V1rj3) that function as cognate receptors for female estrus signals. We also showed that, when combined, the female urine specific cues and estrus signals robustly promote innate male mating behaviors. In this study, we use transgenic approaches to trace vomeronasal sensory neurons (VSNs) expressing V1re and V1rj receptors to elucidate the neural circuitry involved in the processing of female pheromone information. We genetically tagged each receptor gene with a fluorescent reporter or the Cre recombinase. We performed confocal imaging on tissue-cleared accessory olfactory bulbs (AOBs) of adult mice and found stereotypic projection patterns of VSNs. Most of the V1rj-expressing VSNs project to glomeruli concentrated in a distinct spatial location of the anterior AOB (aAOB), whereas glomeruli targeted by V1re-expressing VSNs are more evenly distributed within the aAOB. Glomeruli numbers differ between the V1re and V1rj clades. Furthermore, we found sex differences between glomeruli positions and numbers in AOBs of V1re9-reporter mice. This result suggests that female urine specific cues detected by V1re9-expressing VSNs might be processed in a sexually dimorphic manner. To begin identifying brain regions involved in processing of female pheromone information, we combined the TRAP approach and chemicogenetic tools. Our preliminary findings suggest that sulphated estrogens activate the arcuate nucleus of the hypothalamus (ARH). Additional experiments will provide a model of how the V1re and V1rj receptors act synergistically to promote male courtship behaviors.

Disclosures: T.T. Chiremba: None. **L. Ma:** None. **H. Wilson:** None. **C. McKinney:** None. **Z. Yu:** None. **C.R. Yu:** None.

Poster

PSTR144. Olfaction: Peripheral Mechanisms and Behavior

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR144.20/W17

Topic: D.04. The Chemical Senses

Support: ONR N00014-19-1-2049
ONR N00014-21-1-2343

Title: Investigating the role of OFF neurons in encoding concentration level shifts and odor unsensing

Authors: *F. DENG[1], M. R. TRANER[2], B. RAMAN[3];
[1]Washington Univ. in St Louis, St Louis, MO; [2]Washington Univ. in St louis, St Louis, MO;
[3]Washington Univ. In St. Louis, Washington Univ. In St. Louis, Saint Louis, MO

Abstract: The intensity of a stimulus is a primary encoding dimension for most sensory systems. Both increases and decreases in stimulus intensity are useful information that will help guide an animal toward an odor source or away from toxic cues. However, during trail-following or odor-guided navigation, such level changes have to be detected atop persistent exposure to the odorant. Here, we sought to understand how intensity increments and decrements are encoded in the insect olfactory system. Odorants are detected by sensory neurons in the insect antenna (model organism: locusts; *Schistocera americana*). This information is then transmitted to the antennal lobe neural circuit. Here, cholinergic projection neurons (PNs) and GABAergic local neurons (LNs) interact to transform the sensory input before transmitting this information to higher centers such as the mushroom body (learning and memory center) and lateral horn (putatively drives innate responses). Since the activity of these antennal lobe neurons is the only information available to the higher centers regarding any volatile chemicals in the vicinity of the insects, we examined the responses of the PNs while encountering concentration level shifts. Consistent with our prior results, we found that stimulus introduction and termination activated mostly non-overlapping sets of projection neurons. We found that persistent odor exposure altered the overall activity levels in the antennal lobe network and the ensemble response settled into odor-intensity specific steady-state activity or neural response attractors. Increments and decrements in intensity were encoded by 'opposing' neural ensembles that produced response variations that were inversely correlated with each other. Finally, OFF responses generated after odor cessation encoded information regarding concentration decrements and stimulus termination. Notably, the OFF responses following stimulus cessation were similar irrespective of the concentration of the stimulus terminated (i.e. size of the intensity step-down) indicating that a key role of these OFF responses is to encode the absence of the encountered stimulus.

Disclosures: F. Deng: None. **M.R. Traner:** None. **B. Raman:** None.

Poster

PSTR144. Olfaction: Peripheral Mechanisms and Behavior

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR144.21/W18

Topic: D.04. The Chemical Senses

Support: Stowers Institute for Medical Research
NIH Grant R01DC016696

Title: Regulation of Olfactory Sensory Neuron Plasticity and Axon Targeting by Long Non-coding RNA-H19

Authors: *D. SHALLIE[1], W. XU[1], L. MA[1], C. R. YU[1,2];
[1]Stowers Inst. for Med. Res., Kansas City, MO; [2]Dept. of Anat. and Cell Biol., Univ. of Kansas Med. Ctr., Kansas City, KS

Abstract: Neuronal plasticity in the brain is greatly enhanced early in life, during a time window referred to as the critical period. Mechanisms underlying the heightened plasticity in the olfactory system remain largely unknown. We found that the long non-coding RNA H19 (lncRNA- H19) is expressed in the early postnatal period during olfactory system development. Knockout of H19 did not affect axon targeting but diminished the plasticity during the critical period, preventing the recovery of convergent olfactory sensory axon projection following perturbation. We evaluated the regenerative capacity of olfactory stem cells by tracking the recovery of the olfactory epithelium following ablation by the olfactotoxic drug methimazole and used the incorporation of 5-ethynyl-2'-deoxyuridine to quantitate the rate of proliferation. We observed delayed regeneration and reduced proliferation of the olfactory epithelium in H19 knockout mouse. To understand the change of neonatal circuit plasticity on odor-guided behaviors, we examined odor preference and olfactory imprinting in mice that were genetically altered in the H19 locus. We employed the PROBES system to perform automated single-chamber odor preference assays based on the cross-habituation paradigm. We also developed a threat-induced homing response (TIHR) assay to assess the preference for odorized shelter in response to a looming threat. Consistent with the shortening of the critical period, H19 knockout mice required a shorter odor exposure period to alter innate odor preference when compared to the controls. Result from the TIHR assay further corroborated this finding. Our results reveal a pivotal role of H19 in maintaining the olfactory system and in enabling plastic changes during development.

Disclosures: D. Shallie: None. **W. Xu:** None. **L. Ma:** None. **C.R. Yu:** None.

Poster

PSTR144. Olfaction: Peripheral Mechanisms and Behavior

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR144.22/W19

Topic: D.04. The Chemical Senses

Support: Departmental training grant T32 5T32NS007433
NIH Grant R01DC018516

Title: Defining the role of immature olfactory sensory neurons in olfaction

Authors: *J. D. GREGORY[1,2], R. S. HERZOG[1,2], C. E. CHEETHAM[2];
[1]Neurobio., Univ. of Pittsburgh Grad. Ctr. For Neurosci., Pittsburgh, PA; [2]Dept. of Neurobio., Univ. of Pittsburgh, Pittsburgh, PA

Abstract: To better develop strategies to repair damaged brain areas via stem cell-derived neurons, it is imperative to understand how endogenously generated neurons can functionally

integrate into existing neural circuitry. The mammalian olfactory bulb (OB) is a valuable model to study the functional integration of adult-born neurons in both the healthy and regenerating brain. Adult-born olfactory sensory neurons (OSNs), which are generated throughout a mammal's life, go through immature and mature developmental stages as they wire into the OB. We have shown recently that immature OSNs provide odor input to the mouse OB, where they form monosynaptic connections with excitatory neurons. Furthermore, immature OSNs exhibited graded responses across a wider odorant concentration range than mature OSNs. Therefore, our hypothesis is that immature and mature OSNs provide distinct, but complementary, odor input to OB neurons. To test this, we employ Gg8-tTA;tetO-hM4Di and OMP-IRES-tTA;tetO-hM4Di transgenic mice to chemogenetically silence either immature or mature OSNs respectively via clozapine N-oxide (CNO) mediated activation of the inhibitory DREADD hM4Di. Validation of these models consisted of co-staining olfactory epithelium sections for OMP and HA-tagged hM4Di to determine expression specificity; and phospho-S6 staining of OB sections following odor exposure to quantify the degree of CNO-mediated silencing of OB neurons. Following validation, mice completed both olfactory habituation/dishabituation (odor discrimination) and buried food (odor detection) tests to determine the effect of silencing immature or mature OSNs on odor-guided behaviors. Silencing mature OSNs reduced odor detection and discrimination ability, whereas silencing immature OSNs affected only odor discrimination. Finally, to determine the functional contribution of immature vs. mature OSNs, we imaged odor-evoked responses in GCaMP6s-expressing mitral cells via *in-vivo* 2 photon microscopy before and after hM4Di-mediated silencing. Here we see that silencing either immature or mature OSNs reduced odor-evoked calcium responses in the OB to varying degrees. Together, these experiments provide new insights into the contribution of immature OSNs to odor processing in the healthy OB.

Disclosures: **J.D. Gregory:** None. **R.S. Herzog:** None. **C.E. Cheetham:** None.

Poster

PSTR144. Olfaction: Peripheral Mechanisms and Behavior

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR144.23/W20

Topic: D.04. The Chemical Senses

Support: NIH T32 GM007356
NIMH MH101634
NIMH MH113924
NICHD P50HD103536
NSF 1749772
Cystinosis Research Foundation
Schmitt Foundation

Title: Stability and variability of population activity in the main olfactory bulb across different behavioral states.

Authors: *K. SZYMULA[1,3], K. PADMANABHAN[4,2,5,3];
[1]Biomed. Engin., [2]Ctr. for Visual Sci., Univ. of Rochester, Rochester, NY; [3]MSTP Training Program, [4]Dept. of Neurosci., Univ. of Rochester Sch. of Med. and Dent., Rochester, NY; [5]Univ. of Rochester Intellectual and Developmental Disability Res. (IDDRC), Rochester, NY

Abstract: The rodent olfactory system encodes for odor information through complex spatio-temporal patterns of activity across large populations of mitral and tufted cells, the principal neurons of the main olfactory bulb in rodents. Recent work has demonstrated that the pairwise correlations between mitral and tufted cells differ depending on the animal's behavioral state, for example when it is running versus when it is stationary (Chockanathan et al, 2021). This complements an existing body of literature demonstrating that M/T cell activity and responses to odors can be modulated by behavior (Kay and Laurent, 1999). Thus, a critical question in olfaction remains how the population activity of M/T cells evolves over time, including assessing the stability and variability of neural representations to repeated presentations of volatile odors in the main olfactory bulb (MOB) across different behavioral states. To address this question, we performed high-density recordings (256 channels) by implanting acute arrays into the MOBs of C57BL6 mice (n = 6; 3M/3F) while the animals were head-fixed atop a rotary wheel. Following a period of habituation to different conditions, we examined the activity of large ensembles to an array of 12 odors (that spanned functional group and intrinsic valance) across two different behavioral states (1) locomotion, where the rotary wheel was free to rotate; (2) stationary, where the rotary wheel was locked in place. We found that in addition to complex responses across time in the population to different odors, the behavior influenced the trial-to-trial variability in neural activity. Our preliminary data suggest that understanding how information about odors is encoded for by population activity in the bulb requires an understanding on not only the identity or concentration of the odors, but also the behavioral state of the animal.

Disclosures: K. Szymula: None. **K. Padmanabhan:** None.

Poster

PSTR144. Olfaction: Peripheral Mechanisms and Behavior

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR144.24/W21

Topic: D.04. The Chemical Senses

Support: NIH Grant R01DC017985
NIH Grant R01DC015784

Title: Investigating chemosensation and processing of reptile predator kairomones in the mouse accessory olfactory system

Authors: *J. WANG, J. P. MEEKS;
Dept. of Neurosci., Univ. of Rochester Sch. of Med. and Dent., Rochester, NY

Abstract: Mammalian social interaction relies heavily on chemosensory information processed by olfactory pathways. In terrestrial mammals, the accessory olfactory system (AOS) is specialized for detecting non-volatile cues including kairomones (interspecific social cues) and pheromones (conspecific social cues). Though our knowledge of AOS chemosensation is improving, we still lack knowledge about the majority of natural AOS ligands and the receptors that detect them. Here, we describe studies that investigate the chemosensory mechanisms associated with the detection and perception of threatening kairomones found in predator snake feces. We performed liquid chromatography-mass spectrometry (LC-MS) on the feces of reptilian mouse predators, as well as feces from reptiles with invertebrate or vegetarian diets. These assays revealed that several classes of molecules, including bile acids, metabolic byproducts, dipeptides, and fatty acids, were enriched in the fecal extracts of mouse predators (snakes) compared with fecal extracts from insect-fed lizards and salad-fed turtles. We performed volumetric Ca^{2+} imaging of vomeronasal sensory neurons (VSNs) in the vomeronasal organ (VNO), finding that feces extracts from each class of reptiles elicited distinct activation patterns. This suggests that chemosignals derived from mouse predators activate a distinct population of VSNs. To understand the behavioral impacts of mouse predator kairomones, we applied a machine learning (ML)-based analytic workflow. We quantified mouse behavior from 2D/3D videos of mice interacting with each class of reptilian fecal chemosignals. The results indicate that mice display distinct behavioral patterns to the mouse predator (snake) feces compared to feces from non-mouse-predators. Taken together, this study discovered that a novel population of VSNs is responsible for detecting and processing predatory chemosignals that elicit distinct threat assessment responses in mice. This research will provide the basis for identifying kairomone ligands and corresponding receptors that drive behavioral responses to environmental threats.

Disclosures: J. Wang: None. **J.P. Meeks:** None.

Poster

PSTR144. Olfaction: Peripheral Mechanisms and Behavior

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR144.25/W22

Topic: D.04. The Chemical Senses

Support: KAKENHI, Grant number 20K03482
KAKENHI, Grant number 20K11292

Title: New granule cells in the olfactory bulb are associated with high respiratory input in an enriched odor environment

Authors: S. KAMIMURA[1,2], Y. MASAOKA[3], A. YOSHIKAWA[4], S. KAMIJO[5], ***H. OHTAKI**[6], N. KOIWA[7], M. HONMA[3], K. SAKIKAWA[8], S. KOBAYASHI[2], H. KOBAYASHI[1], T. SHIMANE[1], M. IZUMIZAKI[3];
[1]Otorhinolaryngology Head and Neck Surgery, Sch. of medicine, Showa university, Tokyo,

Japan; [2]Otorhinolaryngology, Sch. of medicine, Showa university Fujigaoka Hosp., Yokohama, Japan; [3]Showa Univ. Sch. of Med., Showa Univ. Sch. of Med., Tokyo, Japan; [4]Showa University, Sch. of Nursing and Rehabil. Sci., Showa University, Sch. of Med., Tokyo, Japan; [5]Dept. of Physiology, Sch. of Pharm., Showa university Sch. of Pharm., Tokyo, Japan; [6]Tokyo Univ. of Pharm. and Life Sci., Hachioji, Japan; [7]Dept. of Hlth. and science, Univ. of Arts and Sci., Saitama, Japan; [8]Showa Univ. Sch. of Med., Showa university Sch. of Med., Tokyo, Japan

Abstract: Olfactory perception largely depends on respiratory activity; olfactory molecules reach the olfactory nerve by inspiration. Respiration plays an important role in initiating sensory transduction in olfactory sensory neurons, and it has been reported respiratory rhythmic activity synchronizes with the bulbar neural circuit. In this study, we assessed whether sniffing activity accompanied by olfactory stimuli is associated with new cells in the olfactory bulb and dentate gyrus of the hippocampus where constantly generated the new neurons. Respiratory activity was continuously measured in mice during exposure to enriched odor stimuli and new cells were stained with 5-bromo-2′-deoxyuridine (BrdU), which selectively labels proliferating cells. Eighteen wild type male mice at 8 weeks of age were randomly divided into two groups: the enriched odor condition and no-odor condition. Two whole-body plethysmographs were placed in a sound-attenuating box, and the plethysmograph measures natural breathing. One chamber was used for the enriched odor condition and the other was used for the no-odor condition. An enriched olfactory environment significantly increased neurogenesis of mitral and granule cells in the olfactory bulb, but not in the dentate gyrus(DG). An increase of new granule cells under the enriched odor condition was correlated to sniffing frequency power, which had a significantly different pattern from the no-odor condition. A high respiratory frequency with frequent odor stimuli may be associated with activation of granule cells to form inhibitory neurons and this active state might increase granule cell neurogenesis. Our study showed that simple odor exposure does not affect hippocampus neurogenesis. Neurogenesis of the DG might be associated with olfactory-associated memory tasks and spatial memory.

Disclosures: S. Kamimura: None. **Y. Masaoka:** None. **A. Yoshikawa:** None. **S. Kamijo:** None. **H. Ohtaki:** None. **N. Koiwa:** None. **M. Honma:** None. **K. Sakikawa:** None. **S. Kobayashi:** None. **H. Kobayashi:** None. **T. Shimane:** None. **M. Izumizaki:** None.

Poster

PSTR144. Olfaction: Peripheral Mechanisms and Behavior

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR144.26/W23

Topic: D.04. The Chemical Senses

Support: NIH Grant R01DC020892 to HL
Subaward to BHS as part of the NSF/CIHR/ DFG/FRQ/UKRI-MRC Next Generation Networks for Neuroscience Program

Title: Mechanosensory Responses in the Antennal Lobe of the Honeybee Apis mellifera

Authors: S. MAHONEY[1], A. ABDALLAH[1], B. SMITH[1], M. BAZHENOV[2], M. PATEL[3], *H. LEI[1];
[1]Arizona State Univ., Tempe, AZ; [2]Univ. of California, San Diego, CA; [3]Col. of William and Mary, Williamsburg, VA

Abstract: Multimodal sensation is a process by which an organism integrates information from multiple sensory modalities to more efficiently process information about a given stimulus and generate the appropriate response. Much work has been done showing the links between different senses, and how they affect one another in the context of dysfunction in this integrative process leading to sensory deficits indicative of a wide range of conditions. However, while the nature of multimodality is well-understood behaviorally, the work on its neurological underpinnings is relatively sparse. The experiment outlined here is the first step of a larger project which aims to use the bimodal integration between olfaction and mechanosensation to better understand how integration occurs at multiple levels of processing. Honeybees are an ideal model for this work, as their antennae are highly multimodal organs, capable of sensing olfactory, mechanosensory, gustatory, and auditory inputs. This experiment establishes a baseline mechanosensory response within the antennal lobe (AL), previously shown to be a major site of integration, using unscented air puffs at various speeds. Using a custom-built olfactometer, we grouped these air flows into different patterns, including continuous puffs at a single air speed, puffs ramping up in speed and then back down, and puffs at randomized speeds. Simultaneously, we performed multichannel extracellular recordings from the AL. Our results show that the spiking rate of AL units is significantly elevated above the baseline firing rate in response to pure mechanosensory stimulation. The magnitude of these responses is proportional to the air speed at which they are presented, with smaller responses to lower speeds and larger responses to higher speeds. As a control, we also stimulated these units with odors, which produced much stronger responses. In summary, we conclude that mechanosensory input can affect odor-evoked response in AL. Experiments that combine the effects of odor concentration and air speed to analyze the interaction between the two are ongoing.

Disclosures: S. Mahoney: None. **A. Abdallah:** None. **B. Smith:** None. **M. Bazhenov:** None. **M. Patel:** None. **H. Lei:** None.

Poster

PSTR144. Olfaction: Peripheral Mechanisms and Behavior

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR144.27/W24

Topic: D.04. The Chemical Senses

Support: F32 MH118724-03S1
1R01 NS123903-01

Title: The olfactory bulb maps breathing rhythms and place in freely-moving mice

Authors: *M. A. BROWN[1], T. M. FINDLEY[1], S. C. STERRETT[2], A. P. WEIBLE[1], M. WEHR[1], J. MURRAY[1], A. L. FAIRHALL[2], M. SMEAR[1];
[1]Univ. of Oregon, Eugene, OR; [2]Univ. of Washington, Seattle, WA

Abstract: Odors carry useful navigational and episodic information, but no matter how many receptor genes are in an animal's genome, there is no receptor for time or place. To optimally orient by olfactory information, brains must unify odor-driven activity with contextual representations of self-movement and -location. Studies in other sensory modalities demonstrate that motor- and location-related signals are common in primary sensory areas. Motivated by these findings, and given the reciprocal connection between olfactory system and hippocampus, we hypothesized that the olfactory bulb encodes contextual information. To test this hypothesis, we captured the sniffing and movement of mice while recording spiking in olfactory bulb (OB), in the absence of experimenter-applied stimuli or tasks. Breathing and spiking differ between head-fixed and freely-moving states. During free movement respiration is rhythmically organized into discrete states lasting minutes, whereas these states are not apparent during head-fixation on a stationary platform. This discrete organization is likewise apparent in the "spontaneous" activity of the olfactory bulb - many individual neurons fire selectively during particular rhythmic states. In addition to these state-selective signals, we also found that allocentric position can be decoded from neuronal ensembles in OB, with comparable decoding performance to hippocampal ensembles recorded under the same conditions. Thus, even during uninstructed behavior and ambient stimuli, contextual information about behavior and place can be read out from the activity of the olfactory bulb. We propose that these contextual signals facilitate the incorporation of olfactory information into cognitive maps of environment and self.

Disclosures: M.A. Brown: None. **T.M. Findley:** None. **S.C. Sterrett:** None. **A.P. Weible:** None. **M. Wehr:** None. **J. Murray:** None. **A.L. Fairhall:** None. **M. Smear:** None.

Poster

PSTR144. Olfaction: Peripheral Mechanisms and Behavior

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR144.28/W25

Topic: D.04. The Chemical Senses

Support: NSF Grant DMS-1547394-004
NIH Grant DC015137

Title: Adult-neurogenesis and the stability-flexibility dilemma in olfactory perceptual memory

Authors: *B. SAKELARIS, H. RIECKE;
Engin. Sci. and Applied Mathematics, Northwestern Univ., Evanston, IL

Abstract: The capacities to learn and store memories are some of the most important functions of the brain, but these two abilities are inherently at odds with each other: a network of neurons must be flexible enough to quickly store new information, but it must be stable enough to

prevent old memories from being overwritten. Olfactory perceptual memory has been shown to be encoded by the olfactory bulb (OB), which is renowned for its large degree of plasticity through adult neurogenesis and structural spine plasticity. This raises the question of how such a highly adaptable system can form stable memories. One possibility implied by theoretical studies in cortical areas is through metaplasticity in which the timescale of synaptic modifications made by an individual neuron changes. We propose that a similar strategy may be implemented in the OB through the exploitation of adult neurogenesis. In the OB, newborn, highly plastic interneurons called granule cells (GCs) consistently arrive in the network throughout adulthood. These neurons have been shown to undergo aging-driven metaplasticity as they exhibit progressively diminished spine turnover over time, potentially allowing them to reconcile the flexibility-stability tradeoff.

Using an anatomically constrained computational model of the OB, we show how neurogenesis, apoptosis, and established properties of GCs, particularly their transiently enhanced excitability and plasticity, combine to enable the flexible formation of stable memories. The model demonstrates that all four of these components are necessary to achieve this goal and to maintain the function of the network as it grows. Moreover, in line with experiments, we show how memories are encoded by young GCs, how these memories are briefly vulnerable to interference from a new stimulus, how re-learning a lost memory is faster than learning a new memory, and how mainly young cells are recruited in simple odor environments, while with increasing complexity, mature cells are also recruited. Furthermore, the model predicts that odor exposure leads to the formation of birthdate-dependent, odor-specific subnetworks in the OB.

Finally, using a simplified mean-field abstraction of our model, we predict that a network with biologically-constrained weights and age-dependent plasticity rates can lead to a memory capacity on the order of $\sqrt{N}$ where N is the number of synapses in the network. This is a vast improvement over the projected capacity of order $\log N$ in a network featuring homogeneous plasticity rates, emphasizing that the augmented capacity of the model predominantly arises from the age-dependent plasticity rates rather than the mere addition of synapses.

Disclosures: B. Sakelaris: None. **H. Riecke:** None.

Poster

PSTR144. Olfaction: Peripheral Mechanisms and Behavior

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR144.29/W26

Topic: D.04. The Chemical Senses

Support: NSF award #2021795

Title: Serotonergic modulation of odor-driven neural and behavioral responses.

Authors: *Y. BESSONOVA, B. RAMAN;
Biomed. Engin., Washington Univ. in St. Louis, St. Louis, MO

Abstract: Olfaction is a fundamental primary sensory modality for many species and it plays a key role in survivaland reproduction. The mapping between olfactory input onto behavioral response often is not fixed, butrather flexible and is regulated by neuromodulatory inputs. Though the role of various neuromodulatorshas been thoroughly explored in the past, how they modulate neural responses in olfactory circuits toproduce variable behavior is poorly understood. In this study, we examined the neuromodulatoryfunction of the widely expressed neuromodulator – serotonin, using *Schistocerca americana* as anexperimental model. Serotonin has been implicated in locusts to trigger phase polymorphism (solitary togregarious phase) which includes rapid and drastic changes in behavior and appearance.We began by characterizing neural responses in the antennal lobe neural network that receivesthe sensory input directly from the antenna. We examined the effects of exogenous serotonin onolfactory responses to different chemical cues. Our result indicates that both spontaneous and odor-evoked responses were altered by the application of serotonin. The spontaneous activity in the antennallobe neurons became more bursty indicating that serotonin altered the excitability of these neurons.Furthermore, odor-evoked responses in individual neurons increased in an activity-dependent manner,and such increases were mostly observed in those neurons that had an initial response to an odorant.While the strength of the odor responses increased (heightened sensitivity), the timing of the neuralresponses and set of neurons activated by an odorant were relatively conserved (stable recognition). Thisresult suggested that serotonin does not modify odor identity but instead increased the sensitivity to it.We used a palp-opening response assay to study the innate behavioral response evoked byodorants. In this assay, locusts open their sensory appendages close to their mouth (called maxillarypalps) upon encounters with a set of appetitive odorants. We found that the application of serotoninincreased the probability of palp-opening responses to an odorant. Notably, such increases in behavioralresponses were not observed for all odorants and were limited to cues that were innately appetitive orneutral. This suggested that serotonin selectively enhanced behavioral responses to odorants, andmatched with the expectations from the physiological datasets. Taken together, this work demonstrateshow serotonin increases the olfactory sensitivity in locusts without altering the original response.

Disclosures: Y. Bessonova: None. **B. Raman:** None.

Poster

PSTR144. Olfaction: Peripheral Mechanisms and Behavior

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR144.30/W27

Topic: D.04. The Chemical Senses

Support: SERB Core Research Grant [CRG/2020/004719]

Title: Understanding the representation of repellents in the antennal lobe of Aedes aegypti

Authors: *K. KANNAN[1], K. ROUT[1], A. BISWAS[1], A. GUPTA[1,2], J. SHUKLA[1,3], S. NAGARKAR JAISWAL[4], N. GUPTA[1];

[1]Dept. of Biol. Sci. and Bioengineering, Indian Inst. of Technol., Kanpur, India; [2]Scholl of Biol. Sci., Univ. of California, San Diego, CA; [3]TATA Inst. for Genet. and Society, Bengaluru, India; [4]CSIR-Centre for Cell. and Mol. Biol., Hyderabad, India

Abstract: Female Aedes aegypti mosquitoes require a blood meal to nourish their developing embryos. They are regarded as a significant hazard due to their capacity to transmit life-threatening diseases to humans and other animals while engaging in blood-feeding activities. DEET is widely regarded as the most efficacious repellent in preventing mosquito bites; however, there remains an ongoing debate regarding the precise mechanism underlying its action. Our findings indicate that in proximity, DEET exhibits independent spatial repellent properties, with Olfactory Receptor Neurons (ORNs) playing a pivotal role in detecting DEET. ORNs and other sensory neurons responsible for olfaction, located in the antenna and maxillary palp of insects, are involved in the detection of external olfactory stimuli. These neurons transmit the sensory information to the antennal lobe (AL), where it synapses onto local neurons (LNs) and projection neurons (PNs) within multiple glomeruli. Following lateral processing by LNs across the glomeruli, PNs convey the information to higher brain centers. For each odor and its respective concentration, a distinct combination of PNs is activated. In order to investigate whether DEET and other aversive odors activate shared glomeruli in the AL, we developed a specific driver line capable of labeling a subset of PNs. By crossing this driver line with a GCaMP reporter line, we effectively induced the expression of GCaMP in PNs. This was validated by observing a significant increase in fluorescence upon the presentation of various odor stimuli, thereby confirming the successful activation of PNs in response to different odors. The findings of this study will provide insights into the representation of repellents in the mosquito brain. Understanding the neural pathways involved in the perception of aversive odors may help in the search for more effective repellents and contribute to the advancement of strategies for vector control.

Disclosures: **K. Kannan:** None. **K. Rout:** None. **A. Biswas:** None. **A. Gupta:** None. **J. Shukla:** None. **S. Nagarkar Jaiswal:** None. **N. Gupta:** None.

Poster

PSTR145. Auditory Processing: Circuit Structure and Function

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR145.01/W28

Topic: D.05. Auditory & Vestibular Systems

Support: HCIR Margaret Messer Student Research Grant
Honors College Undergraduate Research Grant
College of Liberal Arts and Sciences Undergraduate Research Initiative
NIDCD R00 DC019145

Title: Comparative neural architecture in the auditory pathway of echolocating bats

Authors: *N. SUL[1,2], R. MENDOZA[2], V. NIKITIN[4], K. M. BOERGENS[3], A. SALLES[5]; [1]Univ. of Illinois at Chicago, Chicago, IL; [2]Biol. Sci., [3]Physics, Univ. of Illinois Chicago, Chicago, IL; [4]Argonne Natl. Labs, Lemont, IL; [5]Biol. Sci., Univ. of Illinios, Chicago, Chicago, IL

Abstract: Echolocating bats are auditory specialists with highly developed audio-vocal systems that enable navigation using sound. Yet, bats can co-opt these systems and employ them during social interactions. In this way, many bat species have developed wide repertoires of complex communication calls. Although decades of research have focused on auditory processing in bats, there's still much to learn about the neural architecture that supports these specializations. Here, we are aiming to comparatively further the understanding of the architecture of the auditory pathway by exploring the spine density in the auditory cortex (AC), central nucleus of the inferior Colliculus (ICc) and the external nucleus of the inferior colliculus (ICx) across two frugivorous bat species, the laryngeal echolocator *Carollia perspicillata* and a lingual echolocator, *Rousettus aegyptiacus*. Both species use their larynx to produce social vocalizations but adopt very different echolocation strategies and rely differentially on vision to aid navigation. Methods: For this work, the bats were perfused and the brains were extracted, mounted, and cut using a microtome and imaged using a scanning electron microscope (SEM). Prior to the mounting and imaging using the SEM, the sample was sent to Argonne National Laboratory for a synchrotron-based nanoCT scan to image the macroanatomy and ensure staining homogeneity. The images were then aligned and stitched to create a three-dimensional structure for analysis. Analysis was performed by utilizing the software webKnossos which allowed for the tracing of myelinated axons, dendrites and spines up to 13-fold faster than other reconstruction methods. Results: Here, we show for the first time, high resolution electron microscopy images of bat brain tissue and show that methods initially developed for mouse neuroanatomy can be also used across species for comparative research. Furthermore, spine density can be accurately measured in these samples, and our ongoing research focuses on quantifying the differences across areas and species. These findings allow for further comparative work and can be used to identify commonalities and differences across taxa.

Disclosures: N. Sul: None. **R. Mendoza:** None. **V. Nikitin:** None. **K.M. Boergens:** None. **A. Salles:** None.

Poster

PSTR145. Auditory Processing: Circuit Structure and Function

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR145.02/X1

Topic: D.05. Auditory & Vestibular Systems

Support: NIH Grant NS118402
NIH Grant DC020070

Title: A critical period for the synaptic development of octopus cells

Authors: *S. MAEKER[1], L. J. KREEGER[1], L. V. GOODRICH[2];
[1]Harvard Univ., Boston, MA; [2]Neurobio., Harvard Med. Sch., Boston, MA

Abstract: A critical period for the synaptic development of octopus cells
Sydney Maeker, Lauren J. Kreeger, Lisa V. GoodrichOctopus cells (OCs) in the mammalian auditory brainstem are known for their fast membrane properties to the onset of sound and for their large diameter, aspiny dendrites. At hearing onset, OC intrinsic membrane properties and dendritic arbors are morphologically mature. However, their development and synaptic maturation remains poorly understood. Previous work has shown that immature OCs have growth cones and an abundance of filopodia emanating from their dendrites and soma. The functional role that these filopodia play in OC synaptogenesis has never before been characterized. We sought to understand if the filopodia act as a substrate for anatomical plasticity during hearing onset, and if they persist in adult OCs. We made *in vitro* whole-cell recordings in mouse OCs (P9-16, P28) and found activity-dependent intrinsic plasticity can be induced just before the onset of hearing when OCs receive sound-evoked inputs for the first time. We sought to understand if these electrophysiological changes developed in parallel to filopodia variability and synaptic pruning along OC dendrites. To quantify changing morphological properties in developing OCs, we sparsely labeled OCs using the *Thy1-YFP-H* mouse line which allowed us to visualize dendrites and filopodia in detail. We measured filopodia density and length along the dendrites. To determine whether or not OC filopodia had a role in synaptogenesis, we used immunohistochemistry to stain for VGlut1 and Homer1 to determine if and when these filopodia exhibited synaptic machinery. We made 3D reconstructions of whole OCs (P10-14, P30) in Imaris and found that OCs exhibited abundant filopodia at P10, 2 days before hearing onset, and found a sharp decline in filopodia just 2 days post hearing onset. To determine if filopodia present around hearing onset are involved in OC synaptogenesis, we quantified the percentage of filopodia that exhibited pre and postsynaptic partners. Together, our reconstructions and recordings of developing OCs provide insight for the role filopodia play in OC synapse development. We suggest that the onset of auditory input is a critical period for OC development.

Disclosures: S. Maeker: None. **L.J. Kreeger:** None. **L.V. Goodrich:** None.

Poster

PSTR145. Auditory Processing: Circuit Structure and Function

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR145.03/X2

Topic: D.05. Auditory & Vestibular Systems

Support: NIH Grant DC016324

Title: Presynaptic Regulation of Quantal Size at calyx of Held Synapses Following Conductive Hearing Loss

Authors: *T. XIAO, Y. DARWISH, T. WU, H. HUANG;
Tulane Univ., New Orleans, LA

Abstract: In response to a decreased sensory input with hearing loss, the central nervous system undergoes compensatory changes. These changes are associated with neuronal activity alteration and map reorganization, as well as modifications in synaptic function. Using the mouse calyx of Held, a giant glutamatergic terminal in medial nucleus of the trapezoid body (MNTB) of the auditory brainstem, we examined the synaptic function changes following unilateral conductive hearing loss (earplugging). Our results showed that the frequency of miniature excitatory postsynaptic currents (mEPSCs) decreased while mEPSC amplitude increased in hearing loss MNTB compared to the normal side after 2-3 days of earplugging. We also estimated the synaptic properties changes using a high-frequency stimulation protocol, showing a decreased release probability upon hearing loss. Lastly, bath application of a low-affinity, rapidly dissociating AMPA receptor antagonist γ-DGG revealed that higher concentration of glutamate is packed into the vesicles of hearing loss calyces. In conclusion, our results showed that hearing loss induces presynaptic homeostatic plasticity by increasing vesicular glutamate concentration and decreasing neurotransmitter release probability.

Disclosures: **T. Xiao:** None. **Y. darwish:** None. **T. Wu:** None. **H. Huang:** None.

Poster

PSTR145. Auditory Processing: Circuit Structure and Function

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR145.04/X3

Topic: D.05. Auditory & Vestibular Systems

Support: Department of Veteran Affairs RX001095
Department of Veteran Affairs RX001986

Title: Impact of noise exposure and calcium channel blockade on centralneuronal activity and utilization of gap inhibition of acoustic startle reflex as a measure of inferior colliculus function

Authors: *S. YALCINOGLU, S. KOTCHARIAN, R. BRAUN, A. HOLT;
Wayne State Univ. Sch. of Med., Detroit, MI

Abstract: Excessive noise exposure (NE) can induce changes in spontaneous neuronal activity in central auditory pathways, manifesting as hearing loss. Understanding functional changes within central auditory pathways following NE is important for treating noise-induced hearing loss (NIHL). Since voltage-gated calcium channels (CaVs) moderate neuronal activity, they serve as potential therapeutic targets for mitigating NIHL. Thus, the role of verapamil, an L-type calcium channel blocker (CCB), in NIHL was examined in the auditory pathway of rats utilizing auditory brainstem responses (ABRs) and gap inhibition of acoustic startle reflex (ASR_{gi}) temporally following NE.
Male Sprague-Dawley rats were split into four groups: no NE plus saline (n=16), NE plus saline

(n=17), no NE plus verapamil (n=5), and NE plus verapamil (n=7). Verapamil (30 mg/kg) or saline was injected intraperitoneally. The noise groups were unilaterally exposed to a 16 kHz, 106 dB SPL tone for one hour. ABR thresholds and amplitudes of waves I, II, and V were measured at 4, 12, and 20 kHz (0,1,5 days). On the same days on which ABRs were measured, $ASRs_{gi}$ were measured at 4, 8, 12, 16, 20, and 24 kHz. Each group was subjected to a 20 ms startle noise to note maximum startle during silence, in the presence of a background tone, and in the presence of a background tone with a gap prior to startle; the maximum startle response was recorded under each condition.
Verapamil did not affect hearing thresholds in the no noise group but reduced recovery time from the noise-induced temporary threshold shift (TTS). In the noise group, wave I amplitude was less than controls on day 0 of NE ($p<0.007$, 12 and 20 kHz). On day 0, there was trends towards lower wave V amplitude in the noise group compared to controls at 12 and 20 kHz. Prior to NE and treatment, all rats could inhibit their startle. After NE, enhanced ability to inhibit startle response was observed at 12, 16, 20, and 24 kHz ($p<0.05$). Verapamil did not prevent the enhanced ability to inhibit startle response at similar frequencies and was different from controls ($p<0.05$). However, 5 days after treatment, rats in the NE saline group had less ability to inhibit their startle than controls at 16 and 20 kHz ($p<0.05$)
Our results demonstrate that CaVs along the auditory pathway acutely impact noise-induced changes and regulate peripheral and central synaptic function differentially. Since the inferior colliculus (IC) is an important part of the $ASR\ pathway_{gi}$, our results imply that following NE, there is increased excitatory activity within the IC. In the future, the impact of NE and the role of CaVs on central auditory function should be investigated in longitudinal models using ASR_{gi}.

Disclosures: S. Yalcinoglu: None. **S. Kotcharian:** None. **R. Braun:** None. **A. Holt:** None.

Poster

PSTR145. Auditory Processing: Circuit Structure and Function

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR145.05/X4

Topic: D.05. Auditory & Vestibular Systems

Support: NIH grant MH123260
NIH grant GM122645

Title: Corticofugal VIP GABAergic projection neurons in the mouse neocortex

Authors: *A. BERTERO, A. J. APICELLA;
Neuroscience, Developmental and Regenerative Biolgy, UTSA, San Antonio, TX

Abstract: Anatomical and physiological studies have described the neocortex as a six-layer structure that receives, elaborates, and sends out information exclusively as excitatory output to cortical and subcortical regions. This concept has increasingly been challenged by several anatomical and functional studies that showed that direct inhibitory neocortical outputs are also a

common feature of sensory and motor cortices. In particular, the neocortex is characterized by three major classes of inhibitory neurons: Somatostatin, Parvalbumin- and HT3R-expressing neurons. Similarly to their excitatory counterparts, subsets of Somatostatin- and Parvalbumin-expressing neurons have been shown to innervate distal targets like the sensory and motor striatum and the contralateral cortex. However, no evidence of long-range vasoactive intestinal peptide (VIP) neurons, the most represented group among the HT3R-expressing GABAergic cortical inhibitory neurons, has been shown in such cortical regions. Here, using anatomical anterograde and retrograde viral tracing, we tested the hypothesis that mouse cortical VIP-expressing neurons can also send long-range projections to cortical and subcortical areas. We will present data demonstrating, for the first time, that VIP-expressing neurons of the auditory cortex can reach not only the contralateral auditory cortex and the ipsilateral striatum and amygdala, as shown for Somatostatin- and Parvalbumin-expressing expressing long-range neurons, but also the medial geniculate body and both superior and inferior colliculus. We will also present data showing that VIP-expressing neurons of the motor cortex send long-range GABAergic projections to the dorsal striatum and contralateral cortex. Because of its presence in such disparate cortical areas, this would suggest that the long-range VIP projection is likely a general feature of the cortex's network. In addition, by using a combination of *in vivo/ex vivo* whole cell recordings and behavioral paradigms we will present data aiming at understanding the functional role of VIP long-range corticostriatal projection.

Disclosures: **A. Bertero:** None. **A.J. Apicella:** None.

Poster

PSTR145. Auditory Processing: Circuit Structure and Function

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR145.06/X5

Topic: D.05. Auditory & Vestibular Systems

Title: Mechanisms for cancelling self-generated sounds in a cerebellum-like circuit

Authors: *Q. ZHANG, P. CHO, N. SAWTELL;
Neurosci., Columbia Univ., New York, NY

Abstract: The dorsal cochlear nucleus (DCN) integrates auditory nerve input with a diverse array of multimodal signals conveyed by a granule cell-parallel fiber system similar to that found in the cerebellum. While it is hypothesized that the DCN functions to predict and cancel self-generated sounds, the cellular and circuit mechanisms remain unknown. Here we demonstrate that selective optogenetic inhibition of granule cells increases responses to self-generated sounds in putative output neurons of the DCN, while having little impact on auditory tuning. These results, together with recordings from inhibitory cartwheel cells (the main recipients of granule cell input) during feeding behavior, suggest both similarities and differences between mechanisms for cancelling self-generated sensory input in the DCN and those extensively described for cerebellum-like structures in fish.

Disclosures: **Q. Zhang:** None. **P. Cho:** None. **N. Sawtell:** None.

Poster

PSTR145. Auditory Processing: Circuit Structure and Function

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR145.07/X6

Topic: D.05. Auditory & Vestibular Systems

Support: NIH Grant F32MH127788
Kaufman Foundation Grant KA2020-114796
NIH Grant R01DC015527
NIH Grant R01DC014479
NIH Grant R01NS113241

Title: Cortical circuit for integration of auditory and olfactory information

Authors: ***R. CHEN**, N. VOGLER, V. Y. TU, A. C. VIRKLER, J. GOTTFRIED, M. N. GEFFEN;
Univ. of Pennsylvania, Philadelphia, PA

Abstract: In natural environments, animals process sensory stimuli of different modalities simultaneously. Odor and sound are crucial for mice in predator detection and mother-infant interaction. However, we know little about how the brain integrates information of these two modalities. To investigate auditory-olfactory integration, we used viral tracing, immunohistochemistry, and behavioral experiments, and started with a focus on how odors affect sound processing. Anatomically, we identified a direct projection from the piriform cortex (PC) to the auditory cortex (AC) using multiple viral tracers and mouse lines. We first injected retroAAV-hSyn-eGFP in the AC of Cdh23 mice and observed cell-body labeling throughout the PC. Injecting retroAAV-hSyn-Cre in the AC of Ai14 mice yielded consistent results. We quantified labeled cells in different PC subregions and found more cells in the posterior PC projecting to AC compared to the anterior PC. Injections of anterograde AAV1-hSyn-Cre in PC of Ai14 mice showed labeled neurons in all AC subregions, with more labeling in the ventral auditory field. Moreover, we observed cell-body labeling in the auditory thalamus (MGB). To examine the effect of odor on sound perception, we trained mice on a Go/No-Go sound detection task. We observed a decrease in the sound detection threshold on odor-present trials. We took a step further to make both odor and sound stimuli relevant, by training mice on a Go/No-Go task for sound and/or odor detection. We then tested them with combinations of sound and odor stimuli at different intensities. We found that odor modulated sound detection in an intensity-dependent manner. Together, our findings demonstrate integration of auditory and olfactory information and propose a novel pathway enabling such integration.

Disclosures: **R. Chen:** None. **N. Vogler:** None. **V.Y. Tu:** None. **A.C. Virkler:** None. **J. Gottfried:** None. **M.N. Geffen:** None.

Poster

PSTR145. Auditory Processing: Circuit Structure and Function

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR145.08/X7

Topic: D.05. Auditory & Vestibular Systems

Support: NIH Grant SC3NS127766
NIH Grant P30EY013079

Title: Characterization of the thalamic input into monkey auditory cortex, A1

Authors: *V. GARCIA-MARIN[1], M. HAWKEN[2];
[1]Biol., York College/CUNY, Jamaica, NY; [2]Ctr. for Neural Sci., New York Univ., New York, NY

Abstract: The auditory cortex of macaque monkeys is divided into a core of primary areas, surrounded by a narrow belt of associated fields, and a para belt region. The primary core region is further subdivided into A1, R, and RT regions, with A1 receiving a point-to-point thalamo-cortical (TC) input from the medial geniculate (MGv) thalamic nucleus, in a comparable way to the other primary sensory areas S1 and V1. The MGv input accounts for more than 85% of the total thalamic input to A1. Previous studies have used antibodies against an isoform of the glutamate vesicular transporter - VGlut2 - that is only expressed in TC terminals to label the MGv input in macaque A1 and found that it is highly concentrated in the layers IIIb and IV of the core regions. The aim of the current study was to determine the total synaptic density and TC synaptic density in layer 4 of macaque A1 using Transmission Electron Microscopy (TEM) in sections labelled with VGlut2. In addition, we also characterized the ultrastructural properties of the synaptic input in this layer. We found that the total synaptic density was $3.17x10^8$ syn/mm^3 (2.76 and 0.40 syn/mm^3, asymmetric and symmetric, respectively) and the total VGlut2 synaptic density was $0.61x\ 10^8$ syn/mm^3, thus the TC input represents 19% of the total synaptic input and 22% of the asymmetric synaptic input. We measured the ultrastructural characteristics of both asymmetric (As) and symmetric (Sy) synaptic terminals. $Vglut2^+$-As terminals have a greater PSD length (mean 0.325 μm) than non-Vglut2-As terminals (mean 0.278 μm). The Sy terminals (0.254 μm) were smaller than either set of As terminals. Furthermore, the area of the $Vglut2^+$-As-boutons was ~4.5 times larger than the non-VGlut2-As and the Sy terminals (1.725 μm^2, 0.379 μm^2, 0.356 μm^2, for $VGlut2^+$-As, non-vGlut2-As and Sy terminals, respectively). These results confirm that the TC input into the primary sensory areas is stronger than previously thought, as we also demonstrated in macaque V1. In addition, the TC terminals in A1 are much larger in comparison to the intracortical terminals, suggesting that these large terminals could have a stronger synaptic effect in the postsynaptic neurons and contribute to a more effective transmission of feedforward signals.

Disclosures: V. Garcia-Marin: None. **M. Hawken:** None.

Poster

PSTR145. Auditory Processing: Circuit Structure and Function

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR145.09/X8

Topic: D.05. Auditory & Vestibular Systems

Support: STI 2030-Major Projects 2021ZD0200300
NNSFC Grant 31871057, 32070993, 81527901
Beijing Municipal Science & Technology Commission
Z181100001518004, Z181100001518006
Guoqiang Institute Grant

Title: Parvalbumin- and somatostatin-expressing neurons in the inferior colliculus define parallel tectothalamic pathways

Authors: M. LIU[1], Y. GAO[2], T. WANG[3], F. XIN[4], Y. HU[5], ***K. YUAN**[6];
[1]Capital Med. Univ., Beijing, China; [2]Sch. of Med., [3]Sch. of Life Sci., [4]Biomed. Engin., [5]Zhili Acad., Tsinghua Univ., Beijing, China; [6]Biomed. Engin., Tsinghua Univ., Beijing City, China

Abstract: In the ascending auditory pathway, the inferior colliculus (IC) serves as a critical relay between auditory periphery and auditory thalamus (AT). The projections from different subdivisions of the IC to the AT, the so-called tectothalamic pathways, have been thought to be organized in a topographical manner, assuming that neurons in an individual IC subdivision are functionally homogeneous. However, observations that are discrepant from this traditional view have been made, suggesting heterogeneous neuronal functions within an individual IC subdivision. Here, by taking advantage of transgenic mice and various viral tools, we report that the distribution and projection patterns of IC neurons distinctly expressing different molecular markers, parvalbumin (PV) and somatostatin (SOM) in particular, can likely reconcile this discrepancy. Specifically, we identified IC^{PV+} and IC^{SOM+} neurons, which are present in all IC subdivisions, as two of the major cell types in the IC. They predominantly project to the ventral division of the medial geniculate body (MGBv), which is the lemniscal AT, and the posterior limiting nucleus of the thalamus (POL), respectively. The POL is a subdivision of the extralemniscal AT and predominantly projects to the posterior tail of the striatum, which plays a critical role in modulating defensive decision-making. Interestingly, this projection pattern was irrelevant with the specific IC subdivision in which IC^{PV+} or IC^{SOM+} neurons were locally labeled. We further revealed that IC^{PV+} neurons mainly receive inputs from the auditory brainstem, whereas IC^{SOM+} neurons likely integrate processed sensory information of different modalities from various sources. We also demonstrated that IC^{PV+} neurons are more heterogeneous than IC^{SOM+} neurons in terms of their electrophysiological properties, terminal size and neurotransmitter type, likely supporting the robustness of IC^{PV+} neurons in processing acoustic features with great complexity. Our findings provided an explanation for existing discrepancies regarding tectothalamic projections, and suggested a molecular approach for defining tecothalamic pathways in the auditory system.

Disclosures: **M. Liu:** None. **Y. Gao:** None. **T. Wang:** None. **F. Xin:** None. **Y. Hu:** None. **K. Yuan:** None.

Poster

PSTR145. Auditory Processing: Circuit Structure and Function

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR145.10/X9

Topic: D.05. Auditory & Vestibular Systems

Support: NIH K99 DC019415
NIH R01 DC018284
NSF DGE 1256260
NSF 2243919

Title: Neuropeptide Y signaling regulates recurrent excitation in the inferior colliculus.

Authors: ***M. A. SILVEIRA**[1], A. C. DROTOS[1], T. M. PIRRONE[2], T. S. VERSALLE[1], A. BOCK[1], M. T. ROBERTS[1];
[1]Univ. of Michigan, Ann Arbor, MI; [2]Macalester Col., St. Paul, MN

Abstract: Neuropeptides play key roles in shaping the organization, function, and computations of neuronal circuits. We recently found that Neuropeptide Y (NPY), which is a powerful neuromodulator in many brain regions, is expressed in the inferior colliculus (IC) by a distinct class of GABAergic neurons. The IC is localized in a central position in the auditory system, integrating information from numerous auditory nuclei making the IC an important hub for sound processing. In the IC, NPY neurons project locally and send long-range inhibitory projections outside the IC. Previous studies showed that most neurons in the IC have local axon collaterals, suggesting that the IC is rich in local circuits. However, the organization and function of local circuits in the IC remains largely unknown. We previously found that neurons in the IC can express the NPY Y_1 receptor (Y_1R^+) and that application of the Y_1R agonist, [Leu^{31}, Pro^{34}]-NPY (LP-NPY), decreases the excitability of Y_1R^+ neurons. However, how Y_1R^+ neurons and NPY signaling shape local IC circuits is unknown. Here we found that Y_1R^+ neurons represent nearly 80% of glutamatergic neurons in the IC, providing extensive opportunities for NPY signaling to regulate excitation in local IC circuits. Next, to investigate how Y_1R^+ neurons and NPY signaling contribute to local IC circuits, we used optogenetics to activate local Y_1R^+ neurons while recording from other Y_1R^+ neurons as well as neurons that do not express the NPY Y_1 receptor (Y_1R^- neurons). We found that Y_1R^+ neurons provide excitatory input to most other Y_1R^+ and Y_1R^- neurons in the IC and therefore form highly interconnected networks within local IC circuits. Additionally, Y_1R^+ neuron synapses exhibit moderate short-term synaptic plasticity, suggesting that local excitatory circuits maintain their influence over computations during sustained stimuli. We further found that application of LP-NPY decreases recurrent excitation in the IC, suggesting that NPY signaling strongly regulates local circuit function in the

IC. Together, our data show that Y_1R^+neurons are highly interconnected in the local IC and their influence over local circuits is tightly regulated by NPY signaling.

Disclosures: M.A. Silveira: None. **A.C. Drotos:** None. **T.M. Pirrone:** None. **T.S. Versalle:** None. **A. Bock:** None. **M.T. Roberts:** None.

Poster

PSTR145. Auditory Processing: Circuit Structure and Function

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR145.11/X10

Topic: D.05. Auditory & Vestibular Systems

Support: NIH Grant R01DC018353
Nancy Lurie Marks Family Foundation Collaborative Grant

Title: Superficial inhibitory neurons in auditory cortex receive monosynaptic inputs from diverse subregions of the medial geniculate body

Authors: *C. P. MACGREGOR[1], L. G. VATTINO[2,1], C. LIU[3,1], C. G. SWEENEY[2,1], A. E. TAKESIAN[1,2];
[1]Otolaryngology, Massachusetts Eye and Ear Infirmary, Boston, MA; [2]Harvard Med. Sch., Boston, MA; [3]Speech and Hearing Biosci. and Technologies, Harvard Univ., Boston, MA

Abstract: A sparse population of inhibitory interneurons within Layer 1 (L1) of the auditory cortex (ACtx) plays a critical role in auditory plasticity and learning. However, the map of direct inputs onto this population remains incomplete. The ACtx integrates inputs from diverse brain regions, including the medial geniculate body (MGB), the auditory thalamus, which is composed of distinct nuclei that relay both sensory and non-sensory information. Projections from MGB to ACtx primarily target layer 4 (L4), but also extend to cortical L1. Here, we examined the functional and anatomical connections from MGB to two major subtypes of L1 interneurons expressing either vasoactive intestinal peptide (VIP) or neuron-derived neurotrophic factor (NDNF). Using whole-cell electrophysiology recordings during optogenetic activation of MGB afferents, we show that both L1 VIP and NDNF interneurons receive monosynaptic MGB inputs. Furthermore, our results indicate that the MGB-evoked excitatory postsynaptic current (EPSC) amplitudes onto L1 VIP and NDNF interneurons are comparable to those recorded onto L4 excitatory pyramidal neurons, which are known to receive robust MGB input. To examine the distribution of presynaptic neurons amongst the distinct MGB nuclei that provide these monosynaptic inputs to L1 VIP and NDNF interneurons, we employed monosynaptic rabies tracing. The MGB can be subdivided into three major subregions: the primary ventral division (MGBv), and the higher-order medial (MGBm) and dorsal divisions (MGBd), which integrate multimodal sensory and non-sensory information. We found that both L1 VIP and NDNF interneuron populations receive inputs from all three MGB subregions. However, presynaptic neurons that synapse onto VIP interneurons predominantly arise from the MGBv, whereas

neurons synapsing onto NDNF interneurons are more broadly distributed across the primary and higher-order MGB nuclei. These results show that both VIP and NDNF interneurons in superficial ACtx receive monosynaptic inputs from the MGB, but are differentially targeted by distinct nuclei. Together, these findings provide insight into the distinct thalamic inputs that synapse onto L1 interneuron subtypes and motivate future studies that will examine how these subtypes are differentially recruited in vivo during auditory perception and learning.

Disclosures: C.P. MacGregor: None. **L.G. Vattino:** None. **C. Liu:** None. **C.G. Sweeney:** None. **A.E. Takesian:** None.

Poster

PSTR145. Auditory Processing: Circuit Structure and Function

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR145.12/X11

Topic: D.05. Auditory & Vestibular Systems

Title: A multiscale revision of corticothalamic circuit model in the auditory system

Authors: *F. XIE[1], Y. GAO[1], T. WANG[2], **K. YUAN**[3];
[1]Sch. of Medicine,Tsinghua Univ., Beijing, China; [2]Ctr. for Life Sciences, Tsinghua Univ., Beijing, China; [3]Tsinghua Univ., Beijing City, China

Abstract: A multiscale revision of corticothalamic circuit model in the auditory systemAuthors: Fenghua Xie[1,2,3]†, Yixiao Gao[2,3,4]†, Tao Wang[4,5], Kexin Yuan[1,2,3]***Affiliations:** [1] Department of Biomedical Engineering, School of Medicine, Tsinghua University, Beijing 100084, China[2] IDG/McGovern Institute for Brain Research at Tsinghua, Beijing 100084, China[3] Tsinghua Laboratory of Brain and Intelligence (THBI), Beijing 100084, China[4] Center for Life Sciences, Tsinghua University, Beijing 100084, China[5] School of Life Sciences, Tsinghua University†These authors contributed equally: Fenghua Xie, Yixiao Gao* Correspondence should be addressed to Kexin Yuan (kexinyuan@mail.tsinghua.edu.cn)
AbstractLargely topographical projections from different subdivisions of the thalamus, such as the primary, secondary and association sensory thalamus, to hierarchically defined cortical areas have been recognized across sensory systems. However, how corticothalamic projections, which are believed to be crucial for the remarkable flexibility and precision exhibited by our sensory systems, are organized remained poorly understood compared with the thalamocortical counterpart. Here we report that, first, the primary sensory thalamus received direct inputs from cortical L5 neurons. Second, in contrast to the robust thalamocortical topography, L5 neurons in each of the primary, secondary and association auditory cortical areas project to all the subdivisions of the auditory thalamus. Third, the association cortex uniformly provided the most L5 inputs to each individual thalamic subdivisions followed by the secondary auditory cortex. Lastly, L5 axon terminals were mainly varicosity-type and evenly distributed across thalamic subdivisions, but those in the polymodal association subdivisions were the largest. Our data suggest that different subdivisions of the auditory thalamus may under the modulation of

common rather than topographic L5 inputs. The corticothalamic anatomic framework we revealed urges a revisit of current cortico-thalamo-
cortical circuit models in the sensory systems.

Disclosures: **F. Xie:** None. **Y. Gao:** None. **T. Wang:** None. **K. Yuan:** None.

Poster

PSTR145. Auditory Processing: Circuit Structure and Function

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR145.13/X12

Topic: D.05. Auditory & Vestibular Systems

Support: NRF-2021R1A2C3012159
IBS-R002-A2

Title: Modulation of pre-pulse inhibition by the projection from the prefrontal cortex to the inferior colliculus in mice.

Authors: *N. C. D. VAN DER MEEREN[1], **E. JUNG**[2], S.-H. LEE[3];
[1]Biol. Sci., Korean Advanced Inst. of Sci. and Technol. (KAIST), Inst. for Basic Sci. (IBS), Daejeon, Korea, Republic of; [2]Biol. sciences, KAIST, Daejeon, Korea, Republic of; [3]Biol. Sci., Korea Advanced Inst. in Sci. and Technol., Daejeon, Korea, Republic of

Abstract: Sensorimotor gating, a cognitive process, which serves as a mechanism operative in the brain to filter out irrelevant sensory stimuli, prevents excessive load on other brain regions due to repetitive or seemingly less significant sensory information, and suppresses unnecessary motor responses. A well-known paradigm of sensorimotor gating is pre-pulse inhibition (PPI), wherein the brain, exposed to prior sound stimuli, inhibits auditory startle responses in both humans and rodents. While previous research has shed light on the brain regions involved in mediating the PPI, the complete neural circuitry underlying the modulation of PPI remains elusive. In this study, we found that the neuronal projection from the prefrontal cortex (PFC) to the inferior colliculus (IC) plays a key role in modulating the PPI. By conducting anatomical tracing experiments, we have identified direct projections from neurons in the PFC to the IC. Interestingly, when we locally inactivated the PFC through muscimol injection, PPI was enhanced. Moreover, through optogenetic activation of the PFC-to-IC projection coupled with electromyography (EMG) recordings, we observed reduced PPI in wild-type mice, implying the significance of the PFC-to-IC projection in the negative modulation of PPI. Our findings suggest that PFC activity finely tunes the neuronal activity within the IC, and induces hypersensitivity to environmental stimuli. This potentially elucidates the neural mechanism underlying reduced PPI observed in schizophrenia.

Disclosures: **N.C.D. van der Meeren:** None. **E. Jung:** None. **S. Lee:** None.

Poster

PSTR145. Auditory Processing: Circuit Structure and Function

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR145.14/X13

Topic: D.05. Auditory & Vestibular Systems

Support: NIH Training grant DC00046
NIH Grant DC10000

Title: Divergent inhibitory projections of an auditory brain stem nucleus correlate with different intrinsic firing phenotypes

Authors: *J. BALDASSANO[1], K. MACLEOD[2];
[1]Biol., Univ. of Maryland, Col. Park Neurosci. and Cognitive Sci. Program, College Park, MD;
[2]Univ. of Maryalnd, College Park, MD

Abstract: Inhibition has multiple roles in the neural processing of sound at the level of the brain stem in birds. The avian superior olivary nucleus (SON) is the main source of inhibition to auditory brain stem nuclei. SON neurons receive excitatory input from the intensity-coding cochlear nucleus angularis (NA) and the coincidence detectors of nucleus laminaris (NL). SON neurons provide feedback inhibition to the ipsilateral cochlear nucleus magnocellularis (NM), NA, and NL. A separate population of SON neurons projects to the contralateral SON, but whether these two distinct populations differ in their physiological properties is unknown. Studies previously revealed two physiological phenotypes: regular tonic firing (RT) and single spiking (SS). We describe here a third phenotype, a chattering tonic phenotype (CT). In vivo experiments showed that SON neurons have a range of temporal processing capabilities. To determine whether in vitro cell types correlate with postsynaptic target divergence or temporal processing roles, we investigated the circuitry and physiology SON neurons in vitro using patch clamp electrophysiology. First, by electrically stimulating the presynaptic nuclei in specialized wedge slices that contained NA, NL, and SON, we determined that all SON phenotypes could receive excitatory synaptic input from both NL and/or NA, suggesting convergent afferent connectivity. Second, we applied naturalistic fluctuating current injections in vitro that mimicked in vivo inputs. Sensitivity to fluctuations was measured as a change in firing rate, while temporal reliability was assessed using shuffled autocorrelogram analysis. SS neurons were the most sensitive to temporally modulated input and had highest reliability. RT neurons were the least sensitive to temporally modulated input and more closely resembled integrators, while CT neurons had moderate sensitivity and reliability in their firing.Third, intracellular labeling of recorded SON neurons allowed the partial reconstruction of their axonal projections. The projection patterns were strongly related to the physiological phenotypes. SS neurons projected contralaterally, while CT neurons projected ipsilaterally and dorsally in a fiber tract directed toward NM and NL. Finally, the RT neurons projected ipsilaterally via two different fiber tracts, either toward NA or toward NL and NM. These results suggest SON neurons have physiological specializations that allow a range of temporally responsivity, consistent with the diversity of in vivo responses. The data suggests that circuit specializations allow the processing temporal information in functionally distinct pathways.

Disclosures: **J. Baldassano:** None. **K. MacLeod:** None.

Poster

PSTR145. Auditory Processing: Circuit Structure and Function

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR145.15/X14

Topic: D.05. Auditory & Vestibular Systems

Support: NIH/NIDCD R01DC019090
T32DC000011

Title: Auditory cortex differentially modulates inferior colliculus sub-circuits

Authors: *H. M. OBERLE[1], E. J. CHOI[1], C. MARTINEZ-VOIGT[1], P. F. APOSTOLIDES[2]; [1]Univ. of Michigan, Ann Arbor, MI; [2]Univ. of Michigan Med. Sch., Ann Arbor, MI

Abstract: The auditory cortex sends excitatory feedback (corticofugal) projections to the inferior colliculus (IC), a midbrain hub involved in complex sound coding. Corticofugal axons primarily target higher-order dorso-medial and lateral "shell" IC sub-nuclei. We previously characterized corticofugal transmission onto dorsomedial IC neurons, revealing single-cell and network mechanisms that enable powerful non-linear computations in distinct IC cell classes (Oberle et al., 2022; 2023). However, whether corticofugal synaptic activity has similar or divergent effects in the lateral IC is unknown. Here, we combine transgenic mouse lines, optogenetics, and patch-clamp electrophysiology in acute IC brain slices from adult mice. We measured corticofugal transmission in the lateral IC and compared the results with data from dorsomedial IC neurons. We crossed VGAT-ires-cre and Ai14 fl/fl mice to identify GABAergic (VGAT+) and presumptive glutamatergic (VGAT-) neurons during our recordings. We expressed the excitatory opsin Chronos in auditory cortex to optogenetically activate auditory corticofugal axons with trains of light flashes. All dorsomedial IC neurons tested (15/15) exhibited EPSPs during optogenetic stimulation of corticofugal axons; surprisingly fewer lateral IC neurons in the same slices (13/37) showed EPSPs during the same stimulation. Moreover, corticofugal train EPSPs were much smaller in lateral compared to dorsomedial IC neurons, suggesting a sparser convergence of corticofugal axons onto lateral IC neurons. Our prior study (Oberle et al., 2023) showed that corticofugal signals drive polysynaptic excitation in VGAT+ but not VGAT- neurons of the dorsomedial IC. However, preliminary data suggest that this circuit motif is absent in the lateral IC: Onset latencies of corticofugal EPSPs did not significantly differ in VGAT- and VGAT+ neurons. Finally, the lateral IC has a unique organization characterized by GABAergic "modules": Dense clusters of VGAT+ neurons are targeted by somatosensory inputs, but sparse auditory cortex input (Lesicko et al., 2016). By contrast, the surrounding "matrix" zones have a lower density of GABAergic neurons but are densely contacted by auditory corticofugal axons (Lesicko et al., 2016). Interestingly, we find that a number of VGAT+ (4/10) and VGAT- (2/10) cells in the GABA-rich modules responded to optogenetic stimulation of auditory corticofugal fibers despite apparently sparse corticofugal axons in the

vicinity. Future studies will explore the mechanistic bases of this finding. Thus, we identify surprising intricacies of the auditory cortico-collicular pathway's impact on distinct IC sub-regions.

Disclosures: H.M. Oberle: None. **E.J. Choi:** None. **C. Martinez-Voigt:** None. **P.F. Apostolides:** None.

Poster

PSTR145. Auditory Processing: Circuit Structure and Function

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR145.16/X15

Topic: D.05. Auditory & Vestibular Systems

Support: NIH DC004391

Title: Individual cholinergic neurons in the pedunculopontine tegmental and laterodorsal tegmental nuclei send branching axons to innervate the medial geniculate nucleus and ventral tegmental area in guinea pigs

Authors: H. N. SMITH[1], *B. SCHOFIELD[2];
[1]Anthrop., Kent State Univ., Kent, OH; [2]Anat. and Neurobio., Northeast Ohio Med. Univ., Rootstown, OH

Abstract: Cholinergic neurons of the pedunculopontine tegmental nucleus (PPT) and laterodorsal tegmental nucleus (LDT) innervate brainstem and thalamic regions to modulate many neural circuits including limbic, motor and multiple sensory systems. The axons of individual cholinergic neurons frequently branch to innervate multiple target nuclei, allowing a relatively small number of neurons to exert widespread modulation. We have shown previously that PPT and LDT cholinergic neurons innervate the auditory midbrain and thalamus and that individual neurons have axon collaterals that innervate both the inferior colliculus and the medial geniculate (MG) nucleus. However, it is not known if individual cholinergic neurons innervate auditory and non-auditory regions. Here, we ask if cholinergic neurons send axon collaterals to the MG and the ventral tegmental area (VTA), an area involved in encoding "reward" associated with salient stimuli.
We used adult guinea pigs of either sex. We injected different retrograde tracers (red or green Retrobeads, Fluorogold, or Fast Blue) into the MG and the VTA to identify neurons that project to both targets. After 6-9 days, animals were perfused with fixative and the brain sliced into 50 μm sections. We used immunostaining for choline acetyltransferase (ChAT) to identify cholinergic neurons. Cells containing two retrograde tracers were found in the PPT and LDT, indicating that they send collateral projections to both the VTA and MG. In these nuclei, a majority of the double-retrograde cells (27 of 40 cells; 68%; 6 animals) were immunopositive for ChAT (i.e., cholinergic). Other double retrograde cells were ChAT negative; some of these were adjacent to ChAT+ neurons, suggesting that the ChAT-negative cells were glutamatergic or

GABAergic (the other neuronal types present in PPT and LDT). Our data suggest that the cholinergic reward signal sent to the VTA is also sent to the MG, where it could be used to enhance gating of information to the cortex, striatum or amygdala.

Disclosures: H.N. Smith: None. **B. Schofield:** None.

Poster

PSTR145. Auditory Processing: Circuit Structure and Function

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR145.17/X16

Topic: D.05. Auditory & Vestibular Systems

Support: NSTC 111-2423-H-002-012

Title: Decoding neuronal activities in peripheral auditory pathways using DNN models

Authors: P.-T. B. LIU[1,2], Y.-W. LIU[3], Y. TSAO[1];
[1]Res. Ctr. for Information Technol. Innovation, Academia Sinica, Taipei, Taiwan; [2]Dept. of Psychology, Natl. Taiwan Univ., Taipei, Taiwan; [3]Dept. of Electrical Engin., Natl. Tsing Hua Univ., Hsinchu, Taiwan

Abstract: Auditory periphery encodes perceived sounds into spike trains and interacts with the higher auditory system; however, decoding the activities in the auditory periphery remains under-explored in computational neuroscience. In this study, we aim to reconstruct the acoustic stimuli from the spike trains they elicit. To this end, the decoding models must handle the stochastic responses of auditory nerve fibers (ANFs) and compensate for the adaptations by the highly non-linear and bidirectional interactions in the pathways.
The spiking activities were computed by Ray Meddis' Matlab Auditory Periphery (MAP) with the normal hearing condition. The MAP simulated thirty thousand ANFs, which is the number of ANFs in the normal human ear, at high-, medium-, and low-spontaneous firing rates. The ANFs received input from 26 cochlear filters with characteristic frequencies (CFs) between 70 and 8,000 Hz, and the filters were equally spaced on the Equivalent Rectangular Bandwidth (ERB) scale. The MAP also simulated the Acoustic Reflex (AR) and Medial OlivoCochlear Reflex (MOCR) in the efferent pathways. The stimuli consisted of 90 minutes of speech from the LJSpeech dataset and were presented at the 70-dB sound pressure level when computing the ANFs' responses. The spike trains produced by the ANFs were sampled at 22.05k Hz. Thus, the spike trains have limited spectral and relatively good temporal resolutions and were also affected by AR and MOCR adaptations.
Furthermore, we utilized deep artificial neural network-based vocoder models to decode the spike trains of ANFs' responses. There were four training settings based on: 1) different losses, which are L1 and L2, and 2) whether the output waveforms were normalized or not.
The objective metrics were Structural Similarity Index Measure (SSIM) and Mean Square Error (MSE) between the spectrograms of original and reconstructed signals. The first 20 wav files are

in the test set, and the duration is 2 minutes and 12 seconds. The best performance was achieved by the combination of L2 loss and normalized waveforms, with average SSIM and MSE scores of 0.9212 and 0.0028, respectively. The standard deviations of the SSIM and MSE scores were 0.0064 and 0.0006, respectively.
Despite the limited place- and relatively good time-coded information and dynamic adaptations in spike trains, the sounds reconstructed from our models still preserve the speech without noticeable noise or artifacts.
In conclusion, our model reconstructs speech with high fidelity from neuronal spiking activities in human peripheral auditory pathways, and the model can effectively compensate for any nonlinear and dynamical AR and MOCR effects.

Disclosures: **P.B. Liu:** None. **Y. Liu:** None. **Y. Tsao:** None.

Poster

PSTR145. Auditory Processing: Circuit Structure and Function

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR145.18/X17

Topic: D.05. Auditory & Vestibular Systems

Support: NIH Grant R01DC017516
NIH Grant RF1NS128873
Pew Biomedical Scholars
Klingenstein-Simons Fellowship
SFARI Pilot Award
Foundation of Hope

Title: Delineating Parallel Ascending Pathways onto the Secondary Auditory Cortex

Authors: *M. M. GARCIA[1], A. M. KLINE[2], H. TSUKANO[1], C. M. GRAVES[1], P. R. DANDU[1], H. K. KATO[3];
[1]Psychiatry, Univ. of North Carolina at Chapel Hill, Chapel Hill, NC; [2]Psychiatry, Univ. of North Carolina, Chapel Hill, NC; [3]Psychiatry, Univ. of North Carolina Chapel Hill, Chapel Hill, NC

Abstract: How our brain integrates information across parallel sensory channels to achieve coherent perception remains a fundamental question in neuroscience. In the auditory system, sound information reaches the cortex via two parallel pathways. The “primary” lemniscal pathway relays fast and accurate sound information to layer 4 (L4) of the primary auditory cortex (A1). Conversely, the “secondary” non-lemniscal pathway is considered a slow integrator of multisensory information relayed indirectly from cortical areas. Recent anatomical and physiological findings, however, challenge this simple dichotomy. These include our discovery of a short-latency (<10ms) sound input onto layer 6 (L6) of the secondary auditory cortex (A2), comparable in speed to the “fast” lemniscal input to A1 L4. Here, we examined the

hypothesis that this short-latency input is conveyed via non-lemniscal pathways by conducting cortical area- and layer-targeted retrograde tracing. We found that A2 L4 and L6 receive inputs from distinct medial geniculate nucleus (MGN) subdivisions; specifically, A2 L6 receives input from the medial division of MGN (MGm) while A2 L4 is innervated by the caudal part of the ventral division of MGN (MGv). Interestingly, further MGN subdivision-specific retrograde tracing revealed that MGm and caudal MGv receive inputs from overlapping but distinct domains of the shell of the inferior colliculus, which in turn receive direct input from the cochlear nucleus. These findings demonstrate a non-lemniscal origin of parallel ascending pathways that bypass A1 and directly reach both the superficial and deep layers of A2. Moreover, our results suggest that caudal MGv, but not rostral MGv, belongs to the non-lemniscal pathway, despite the conventional view of MGv as a homogeneous lemniscal structure. Ongoing electrophysiology and optogenetic manipulation studies aim to investigate the sound response properties of these non-lemniscal pathways and explore how parallel ascending pathways are integrated in the cortex to shape perception.

Disclosures: **M.M. Garcia:** None. **A.M. Kline:** None. **H. Tsukano:** None. **C.M. Graves:** None. **P.R. Dandu:** None. **H.K. Kato:** None.

Poster

PSTR145. Auditory Processing: Circuit Structure and Function

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR145.19/X18

Topic: D.05. Auditory & Vestibular Systems

Support: NIH Grant R01DC017516
NIH Grant RF1NS128873
Pew Biomedical Scholars
Klingenstein-Simons Fellowship
SFARI Pilot Award
Foundation of Hope

Title: Predictive filtering of primary auditory cortex activity by frontal top-down inputs

Authors: *H. TSUKANO, M. M. GARCIA, P. R. DANDU, C. M. GRAVES, H. K. KATO; Dept. of Psychiatry, Univ. of North Carolina at Chapel Hill, Chapel Hill, NC

Abstract: Our brains continuously compare incoming sensory inputs against predictions based on previous experiences, assigning less salience to predictable stimuli. While sensory habituation to repetitive stimuli is considered the simplest manifestation of this predictive coding, the circuit mechanisms underlying long-term habituation over days remain unclear. We previously reported that daily passive sound exposure attenuates neural responses in the mouse primary auditory cortex (A1), a plasticity mediated by local inhibition from somatostatin-expressing neurons (SST neurons). In the current study, we further explored the source of top-down predictive signals

regulating SST neurons to create a “negative image” that cancels out sound input. Retrograde tracing demonstrated that A1 SST neurons receive projections from frontal cortical areas, most prominently from the orbitofrontal cortex (OFC). Two-photon calcium imaging of OFC axon terminals in A1 revealed enhanced top-down input following daily passive tone exposure, supporting its role in encoding predictive signals. Muscimol infusion into the OFC reversed the pre-established habituation in A1 by enhancing pyramidal neuron activity while suppressing SST neuron activity. Importantly, this effect was absent in naïve animals, highlighting the specific involvement of the OFC in experience-dependent predictive filtering. We also found that the deletion of NMDA receptors in SST neurons reduces habituation, pointing to the role of synaptic plasticity at inputs onto SST neurons. Together, our findings suggest that the predictive filtering of sensory activity is realized by a global circuit mechanism recruiting the frontal top-down inputs.

Disclosures: H. Tsukano: None. **M.M. Garcia:** None. **P.R. Dandu:** None. **C.M. Graves:** None. **H.K. Kato:** None.

Poster

PSTR145. Auditory Processing: Circuit Structure and Function

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR145.20/X19

Topic: D.05. Auditory & Vestibular Systems

Support: NIH Grant R01DC017516
NIH Grant RF1NS128873
Pew Biomedical Scholars
Klingenstein-Simons Fellowship
SFARI Pilot Award
Foundation of Hope
Toyobo Biotechnology Foundation
Japan Society for the Promotion of Science Overseas Research Fellowship

Title: Inter-areal cortical circuits underlying the extraction of complex acoustic features

Authors: *K. ONODERA[1], A. M. KLINE[2], H. K. KATO[3];
[1]Dept. of Psychiatry, Univ. of North Carolina Chapel Hill, Chapel Hill, NC; [2]Dept. of Psychiatry, Univ. of North Carolina, Chapel Hill, NC; [3]Dept. of Psychiatry, Univ. of North Carolina at Chapel Hill, Chapel Hill, NC

Abstract: The auditory cortex consists of primary and higher-order regions interconnected to form hierarchical streams for sound information processing. Despite ample evidence suggesting the critical roles of higher-order auditory cortices in extracting complex acoustic features, how they interact with primary areas for their unique computations remains unclear. Here, we used area-specific optogenetics in mice to investigate the role of inter-areal circuits in sound

processing within individual regions. We began by re-evaluating the hierarchy among the primary auditory cortex (A1), anterior auditory field (AAF), and secondary auditory cortex (A2), given recent studies questioning A2's classification as a higher-order cortex. Optogenetic silencing of one region while simultaneously recording from the other two revealed reciprocal excitation primarily in the superficial layers of all area pairs. A1→A2 showed the most robust excitation, followed by A2→A1 and A1↔AAF connectivities. Surprisingly, minimal interaction was observed between AAF and A2, despite their geographical proximity. Additional analyses of noise correlation and Granger causality between area pairs validated these observations. Our results thus support a hierarchy between A1 and A2, while AAF appears to form a distinct information processing stream. Building on the significant mutual interaction between A1 and A2, our ongoing research investigates how these two areas cooperate in extracting complex acoustic features. Our prior studies identified preferential representations of frequency-modulated sweeps in A1 and coincident harmonics in A2. Leveraging these sound features, we examine how A1 and A2 cooperate to construct unified representations for sounds with combined features, such as frequency-modulated harmonics. By elucidating the dynamic interplay between primary and secondary cortices, our study aims to decipher the principles underlying the extraction of complex features within hierarchical sensory streams with recurrent connections.

Disclosures: **K. Onodera:** None. **A.M. Kline:** None. **H.K. Kato:** None.

Poster

PSTR145. Auditory Processing: Circuit Structure and Function

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR145.21/X20

Topic: D.05. Auditory & Vestibular Systems

Title: Global activity in auditory cortical circuits counteracts sparse optogenetic inhibition near-instantaneously

Authors: *R. STEINFELD, W. F. PODLASKI, C. K. MACHENS, A. RENART; Champalimaud Fndn., Lisboa, Portugal

Abstract: Brain functioning displays remarkable robustness, including against optogenetic interventions that lead to rapid inhibition of activity in affected neurons. However, a systematic study of how the dynamics of cortical circuits respond to such perturbations has not yet been conducted. Here, we addressed this problem by recording the responses of large populations of neurons in the auditory cortex of awake mice in response to partial silencing of neurons in the circuit. First, we optimized the expression level of the inhibitory opsin stGTACR2 to induce silencing in a sparse subset of auditory cortical neurons by shining blue light with an LED implanted above a cranial window. Using multi shank Neuropixels 2 probes, we then recorded population activity in response to a set of artificial vowels. We report that auditory cortical circuits are able to quickly and actively counteract the induced perturbation across various

degrees of inhibition within the population. While a fraction of neurons - which depends on the density of the opsin's expression - was profoundly inhibited by light, the sound responses of most non-inhibited neurons increased in trials with optogenetic perturbation. As a result, the global activity during sound presentation, calculated across all recorded units and experiments, was only modified by 9% in LED trials, whereas the average absolute (unsigned) change in activity caused by the LED was 40%. In addition to this approximate preservation of the total activity level of the neural sound responses, the shape of the average time-varying response was also remarkably conserved. Our results uncover a striking capacity of cortical circuits for near-instantaneous compensation, in line with predictions of balanced spiking networks or cooperative neural codes.

Disclosures: R. Steinfeld: None. **W.F. Podlaski:** None. **C.K. Machens:** None. **A. Renart:** None.

Poster

PSTR146. Photoreceptors and Retinal Circuitry

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR146.01/X21

Topic: D.06. Vision

Support: NIH Grant DC012938

Title: Quantal analysis of glycine release at the inhibitory synapses of a retinal amacrine cell

Authors: *P. STRAZZA JUNIOR, H. VON GERSDORFF;
Vollum Inst., Oregon Hlth. and Sci. Univ. (OHSU), Portland, OR

Abstract: The AII amacrine cells (AII-ACs) play a central role in retinal crossover inhibition where they convert excitatory light responses from Rod Bipolar (RBCs) and ON-Cone Bipolar (ON-CBCs) cells into glycinergic inhibition of OFF-Cone Bipolar cells (OFF-CBCs). Recently, our group have shown that synaptic vesicle exocytosis by AII-ACs is potentiated by cAMP through a signaling pathway relying on EPAC activation and Ca^{2+} release from internal Ca^{2+} stores, but the mechanisms initiating this potentiation of glycine release remain elusive. To investigate the mechanisms that control glycinergic neurotransmission between AII-ACs and OFF-CBCs in adult mouse retinal slices, we performed patch clamp recordings from OFF-CBCs using a Syt2-GFP mouse line that labels OFF-CBCs. During drug washes we performed whole-cell recordings of presynaptic AII-AC membrane potential and spiking (current-clamp) and postsynaptic OFF-CBC glycinergic spontaneous IPSCs (sIPSCs; voltage-clamp). We found that the Na^{+} channel blocker TTX and also L-AP4, a blocker of RBC and ON-CBC synaptic inputs, hyperpolarized the AII-ACs. Voltage clamp experiments in OFF-CBCs revealed that both L-AP4 and TTX decrease the frequency and average amplitude of the sIPSCs. We next isolated single glycinergic vesicle fusion events (quantal events) by blocking Ca^{2+} channels with cadmium (Cd^{2+}) or by blocking L-type Ca^{2+} channels with isradipine and Na^{+} channels with TTX.

Statistical analysis of noise, single quanta and multiquantal events revealed clear multi-Gaussian peaks, with mean amplitude values linearly distributed. Histograms of sIPSC amplitudes with a single peak around 25 pA were present in half of the recordings with Ca^{2+} channel blockers. Analysis of sIPSCs risetimes revealed that larger events had on average slower risetimes. Together these results suggest that larger sIPSCs represent highly synchronous multiquantal events reflecting the fusion of multiple synaptic vesicles. Our analysis shows that the membrane potential of AII-ACs controls the probability of glycine release in a rapid, graded and sustained manner. These findings will contribute to a better understanding of how the retina adapts crossover inhibition circuits during changes in environmental luminance and contrast.

Disclosures: P. Strazza Junior: None. **H. von Gersdorff:** None.

Poster

PSTR146. Photoreceptors and Retinal Circuitry

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR146.02/X22

Topic: D.06. Vision

Support: NIH R01EY019498
NIH R01EY013528
Society of Hellman Fellows
NIH R00EY028625

Title: Single-cell analysis reveals weak influence of cholinergic retinal waves on the gene expression states of retinal ganglion cell types

Authors: *R. SOMAIYA, M. A. PO, K. SHEKHAR, M. B. FELLER;
Univ. of California, Berkeley, Berkeley, CA

Abstract: In the early stages of retinal development, a form of correlated activity known as retinal waves causes periodic depolarizations of immature retinal ganglion cells (RGCs). Retinal waves are crucial for refining visual maps in the brain's retinofugal targets as well as development of retinal circuits, including direction selectivity. However, whether these waves alter gene expression levels in RGCs has not been studied. We performed single-cell RNA sequencing on RGCs isolated from control mice and β2KO mice, which lack the β2 subunit of the nicotinic acetylcholine receptor, resulting in the disruption of retinal waves. Using computational methods, we compared β2KO RGCs to control RGCs to identify gene expression changes resulting from the disruption of waves. Our results suggest a surprisingly weak impact of retinal waves on gene expression levels of RGCs, both globally and at the level of molecularly and functionally defined types. Thus, it is unlikely that retinal waves influence the cell-intrinsic genetic or epigenetic programs that instruct RGC differentiation and maturation. Notably, among the few genes significantly impacted in β2KO RGCs was *Kcnk9*, which encodes the two-pore

domain pH-sensitive potassium channel TASK3. Here, we will present validation of this result using both *in situ* hybridization and functional characterization of this conductance.

Disclosures: **R. Somaiya:** None. **M.A. Po:** None. **K. Shekhar:** None. **M.B. Feller:** None.

Poster

PSTR146. Photoreceptors and Retinal Circuitry

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR146.03/X23

Topic: D.06. Vision

Support: a career development award from Research to Prevent Blindness
a career starter award from Knights Templar Foundation
Klingenstein-Simons neuroscience fellowship
an unrestricted grant to the Department of Ophthalmology from Research to Prevent Blindness

Title: Molecular characterizations of retinal cell types in the sea lamprey

Authors: *J. WANG, L. ZHANG, A. PAHLEVAN, J. SUN, M. CAVALLINI, A. MORSHEDIAN, G. FAIN, A. P. SAMPATH, Y.-R. PENG;
Dept. of Ophthalmology, Stein Eye Institute, David Geffen Sch. of Medicine, Univ. of California Los Angeles, Los Angeles, CA

Abstract: The origins of retinal cell types, as well as their diversification across vertebrate species, remain elusive. As an ancient jawless vertebrate species, the lamprey offers an important model to probe the evolutionary history of retinal cells. In this study, we generated a cell atlas of the adult sea lamprey retina using single-cell RNA sequencing and provided a computational framework to compare the lamprey retinal cell types with those found in jawed species. Firstly, we identified six conserved retinal cell classes: photoreceptor cell (PR), horizontal cell (HC), bipolar cell (BC), amacrine cell (AC), retinal ganglion cell (RGC), and Müller glia (MG). We also validated these classes using in situ hybridization (FISH). Furthermore, we identified 2 PR, 6 HC, 6 BC types, 19 AC, 41 RGC, and 4 MG types. Secondly, to uncover master regulators that specify cell classes, we employed network-based methodologies (ARACNe-AP and Rosa) to infer protein activity. We identified class-specific regulators, including transcription factors (TFs), transcription cofactors (coTFs), and cellular surface proteins. We next evaluated the conservation of these regulators across species by performing protein set enrichment analysis. We found that the conservation varies across cell classes with MG exhibiting the highest degree of conservation and HC the lowest. Finally, we reconstructed the diversification history of RGC types by conducting a comparative analysis of transcriptomic data from lamprey, mouse, and macaque species. We found that RGCs had already diversified to distinct subsets of types in a common ancestor shared between the lamprey and jawed vertebrate species, while species-specific RGC types are generated superimposed on these subsets. Therefore, our findings provide

valuable insights into understanding the essential regulators responsible for maintaining cell class identities and shed light on the evolutionary history of the specification of retinal cell types.

Disclosures: **J. Wang:** None. **L. Zhang:** None. **A. Pahlevan:** None. **J. Sun:** None. **M. Cavallini:** None. **A. Morshedian:** None. **G. Fain:** None. **A.P. Sampath:** None. **Y. Peng:** None.

Poster

PSTR146. Photoreceptors and Retinal Circuitry

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR146.04/X24

Topic: D.06. Vision

Support: NINDS Intramural Reasearch Program NS003145

Title: En passant bipolar cell synaptic inputs to dopaminergic amacrine cells in mouse retina

Authors: *A. NATH[1], K. P. SZATKO[1], D. M. BERSON[2], J. S. DIAMOND[1];
[1]NINDS, NIH, Bethesda, MD; [2]Dept. of Neurosci., Brown Univ., Providence, RI

Abstract: In the mammalian retina, dopamine is synthesized in and released from dopaminergic amacrine cells (DACs). Previous studies show that DACs receive excitatory inputs at light onset[1-5] but their dendrites mostly stratify in the OFF sublamina of the inner plexiform layer (IPL)[3,6,7]. Close examination of a serial electron microscopic (EM) data set acquired from mouse retina revealed that DACs receive majority of their excitatory synaptic inputs from *en passant* synapses made by type 6 and type 9 CBCs in the accessory ON layer of IPL. Type 6 CBCs also make substantial gap junction connections with AII ACs in the ON layer[8] and thereby may enable highly sensitive scotopic signals from the primary rod pathway to flow to DACs.We performed whole cell voltage clamp recordings of DACs in Drd2-GFP mice and presented brief flashes from darkness to investigate sensitivity of DAC excitatory inputs. Flash stimuli elicited weak excitatory postsynaptic currents (EPSCs) that reached maximal amplitude at ~25,000 R*/rod/s. Blocking synaptic inhibition using a cocktail of strychnine, gabazine and TPMPA increased EPSC amplitudes and significantly decreased (i.e., by ~50-fold) the light intensity required to elicit a half-maximal response. Sensitization of DAC responses via $GABA_A$, $GABA_C$ and glycine antagonists was also reflected in DAC membrane voltages measured under current clamp. These results suggest that scotopic signals are prevented from entering the accessory ON layer via shunting inhibition imposed by amacrine cells (ACs) onto type 6 CBC axons near the *en passant* synaptic contacts with DACs. EM analysis of ACs providing inhibitory inputs to type 6 CBCs suggest MAC[9] and bistratified VIP AC[10,11] to be the likely candidates.

References

1.Zhang, D.-Q., Stone, J. F., Zhou, T., Ohta, H. & McMahon, D. G. *Neuroreport* 15, 1761-1765 (2004).
2.Prigge, C. L. *et al. J Neurosci* 36, 7184-7197 (2016).
3.Newkirk, G. S., Hoon, M., Wong, R. O. & Detwiler, P. B. *J Neurophysiol* 110, 536- 552

(2013).
4.Qiao, S.-N., Zhang, Z., Ribelayga, C. P., Zhong, Y.-M. & Zhang, D.-Q. *Sci Rep-uk* 6, 28916 (2016).
5.Zhao, X., Wong, K. Y. & Zhang, D.-Q. *Sci Rep-uk* 7, 7920 (2017).
6.Gustincich, S., Feigenspan, A., Wu, D. K., Koopman, L. J. & Raviola, E. *Neuron* 18, 723-736 (1997).
7.Witkovsky, P. *Doc Ophthalmol* 108, 17-39 (2004).
8.Tsukamoto, Y. & Omi, N. *Front Neuroanat* 11, 92 (2017).
9.Grimes, W.N. *et al.* A high-density narrow-field inhibitory retinal interneuron with direct coupling to Müller glia. *J. Neurosci*. 41, 6018-6037.
10.Zhu, Y., Xu, J., Hauswirth, W. W. & DeVries, S. H. *J Neurosci* 34, 7845-7861 (2014).
11.Park, S. J. H. *et al. J Neurosci* 35, 10685-10700 (2015).

Disclosures: A. Nath: None. **K.P. Szatko:** None. **D.M. Berson:** None. **J.S. Diamond:** None.

Poster

PSTR146. Photoreceptors and Retinal Circuitry

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR146.05/X25

Topic: D.06. Vision

Support: CIHR project grant
NSERC discovery grant

Title: Cadherin 4 directs the assembly of a family of retinal circuits that encode unique aspects of dark visual stimuli

Authors: *A. RANGEL OLGUIN[1], P.-L. ROCHON[2], C. THERIAULT[1], A. KRISHNASWAMY[1];
[1]Physiol., McGill University, Montreal, Montreal, QC, Canada; [2]Integrated Program in Neurosci., McGill Univ., Montreal, QC, Canada

Abstract: Retinal ganglion cells (RGCs) must grow dendrites into a specific sublamina of the inner plexiform layer (IPL) to synapse with appropriate retinal interneurons and become selective for a unique visual feature such as motion, edges, etc. The molecular determinants of this important event are not entirely clear; however, our previous work suggests that members of the cadherin (Cdh) superfamily may play a critical role. By relating differentially expressed Cdhs in RGCs to their dendritic patterns, we assigned each of the 5 IPL sublamina as being positive for a specific combination of 8 Cdhs. Using histological stains and Cre-ER knockin lines, we focused on one of these candidates, Cdh4, and described that it labels a family of RGCs that targets the outer IPL sublaminae. Using dense calcium imaging methods and posthoc immunostaining, we found that Cdh4-RGCs comprise 8 RGC types and showed that each prefers a unique aspect of dark visual objects. We hypothesized that this dark object preference arises

from their sublamina-specific targeting. To test this hypothesis, we repeated these studies in the absence of Cdh4 and observed that C4-RGCs fail to target appropriate sublamina and exhibit major deficits in stimulus selectivity. These results show that Cdh4 directs RGC dendrites to outer sublaminae so they can become selective for dark visual objects, and strongly suggests that Cdh expression in retinal neurons acts as a molecular blueprint for circuit assembly.

Disclosures: A. Rangel Olguin: None. **P. Rochon:** None. **C. Theriault:** None. **A. Krishnaswamy:** None.

Poster

PSTR146. Photoreceptors and Retinal Circuitry

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR146.06/Y1

Topic: D.06. Vision

Support: Max Planck Society
HFSP

Title: Visual microcircuitry of the cuttlefish Sepia officinalis

Authors: *D. EVANS[1], A. KARIMI[2,1], G. LAURENT[1];
[2]Dept. of Connectomics, [1]MPI Brain Res., Frankfurt am Main, Germany

Abstract: The optic lobe of the cuttlefish *Sepia officinalis* receives direct retinal photoreceptor input and processes visual information about textures in order to generate camouflage patterns. We employed serial block-face electron microscopy to acquire 3D-EM volumes (sized 140 x 200 x 200 μm^3 at a resolution of 11 x 11 x 30 nm^3) from the cortical optic lobe of juvenile and adult animals. As this is the site of the first optic synapse of cephalopod photoreceptors, we are reconstructing their postsynaptic targets to identify early connectivity motifs in the cephalopod visual system, and ultimately aim to link these to functional types. This work aims to understand the connectome of early visual circuits in *Sepia officinalis*.

Disclosures: D. Evans: None. **A. Karimi:** None. **G. Laurent:** None.

Poster

PSTR146. Photoreceptors and Retinal Circuitry

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR146.07/Y2

Topic: D.06. Vision

Support: Max Planck Society
HSFP

Title: Cellular characterisation of functional subtypes in the early visual system of sepia officinalis

Authors: *A. KARIMI[1,2], D. EVANS[3], G. LAURENT[3];
[1]Neural Systems, MPI Brain Res., Frankfurt, Germany; [2]Connectomics, [3]Neural Systems, Max Planck Inst. for Brain Res., Frankfurt am Main, Germany

Abstract: Visual textures are processed to generate camouflage skin patterns in the cuttlefish Sepia officinalis. The optic lobe, directly innervated by photoreceptors and as few as two synapses upstream from the skin's chromatophores, is the prime candidate for this transformation of visual scene statistics into complex behaviour. It comprises a retinorecipient cortex of two granule cell layers and a plexiform layer reminiscent of the cytoarchitecture of vertebrate retina, and a sub-cortical medulla where cells are organised in a fractal tree. Using two-photon imaging of calcium dyes in the optic lobe of Sepia hatchlings, we characterised the tuning properties of optic lobe cells in response to visual stimulation. We find the presence of ON, OFF and ON-OFF cells in all layers of the cortex, with a diversity of temporal tuning profiles described in vertebrate and non-vertebrate retinas. This study aims to identify functional subtypes in a visual system with a camera-type eye that has evolved through convergence.

Disclosures: **A. Karimi:** None. **D. Evans:** None. **G. Laurent:** None.

Poster

PSTR146. Photoreceptors and Retinal Circuitry

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR146.08/Y3

Topic: D.06. Vision

Support: KAKENHI 22KK0137
KAKENHI 19H01140
KAKENHI 24390019
Kobayashi Foundation

Title: Mechanism of the oscillation in retinal ganglion cells of Trpm1 KO mouse

Authors: *S. HORIE[1], K. SAKUTA[2], K. TADA[3], K. KITANO[4,5,6], M. TACHIBANA[6], C. KOIKE[2,5,6];
[1]Grad. Sch. of Pharm., [2]Col. of Pharmaceut. Sci., [3]Grad. Sch. of Information Sci. and Engin., [4]Col. of Information Sci. and Engin., [5]Res. Organization of Sci. and Technology, R-GIRO, [6]Res. Organization of Sci. and Technology, Ctr. for Systems Vision Sci. (CSVS), Ritsumeikan Univ., Shiga, Japan

Abstract: Light-evoked responses of retinal ON-type bipolar cells (BC) are generated by transient receptor potential cation channel subfamily M member 1 (Trpm1), which is regulated by metabotropic glutamate receptor subtype 6 (mGluR6). It has been reported that abnormal periodic spontaneous firing (oscillation: 3 - 8 Hz) is observed in retinal ganglion cells (RGCs) of Trpm1 knockout (KO) mice. Here, we investigated how the oscillation is generated in RGCs. Using whole-mount retinal preparation of 1-month-old Trpm1 KO mice, we applied the patch-clamp technique to alpha RGCs (αRGCs). The electrode solution contained a Cs-based solution with QX 314 and neurobiotin. After recording, the retina was immunostained with ChAT and SMI-32 antibodies, and the recorded cell was morphologically classified into ON and OFF types based on the dendritic stratification in the inner plexiform layer. We checked the light-evoked firing and confirmed the oscillatory spike discharges in the cell-attached mode. Then, the cell was whole-cell voltage-clamped at -60 mV near the resting membrane potential. Under this condition, we observed periodic synaptic inputs, showing that the oscillation is generated presynaptically. To examine whether the synaptic input is excitatory or inhibitory, the reversal potential of the periodic synaptic current was estimated. We found the reversal potential near the Cl^- equilibrium potential and 0 mV in OFFαRGCs and ONαRGCs, respectively. The oscillation may likely be driven by glycine/GABAergic inputs in OFFαRGCs and glutamatergic input in ONαRGCs. Next, we examined whether the phase of oscillation is the same or different between nearby αRGC pairs. It was found that the oscillation was in-phase in ON/ON or OFF/OFF pairs, whereas it was antiphase in ON/OFF pairs. These results suggest that oscillatory presynaptic cells may simultaneously send inhibitory and excitatory inputs to the OFF and ON pathways in the inner plexiform layer, respectively. We assume that the oscillatory cell may be AII amacrine cells (AII ACs), which receive glutamatergic inputs from rod (ON) BCs and form gap junctions with ON cone BCs as well as glycinergic synapses with OFF cone BCs and some OFF RGCs. Simulation with a retinal circuit model estimates that a slight hyperpolarization of AII ACs generates the oscillation, which is transmitted to ON and OFF RGCs. We have observed that the rod BC terminal is reduced in size in Trpm1 KO mice, and thus AII ACs may hyperpolarize due to decreased excitatory inputs from rod BCs. In conclusion, the oscillation observed in RGCs of Trpm1 KO mouse is generated by synaptic inputs from oscillatory cells (probably AII ACs).

Disclosures: **S. Horie:** None. **K. Sakuta:** None. **K. Tada:** None. **K. Kitano:** None. **M. Tachibana:** None. **C. Koike:** None.

Poster

PSTR146. Photoreceptors and Retinal Circuitry

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR146.09/Y4

Topic: D.06. Vision

Support: NIH Grant EY031677
McPherson Eye Research Institute

Title: Role of inhibition in shaping ON-alpha retinal ganglion cell function

Authors: *A. SAWANT[1], R. SINHA[2], M. HOON[3];
[1]Univ. of Wisconsin Madison, Madison, WI; [3]Ophthalmology and Visual Sci., [2]Univ. of Wisconsin, Madison, Madison, WI

Abstract: Outputs of neural circuits, such as the retina, are determined by an interplay of excitatory and inhibitory signals. In the retinal output neurons (ganglion cells), synaptic excitation from presynaptic neurons (bipolar cells) integrates with synaptic inhibition from interneurons (amacrine cells) to regulate ganglion cell output. For the well characterized ON-alpha retinal ganglion cell (ONa RGC) circuit, which is involved in luminance and dim light encoding, we know a lot about the role of excitation in shaping functional output, but little is known about the role of inhibition and how it integrates with excitation to regulate ONa spike output. ONa RGC dendrites express both GABA and Glycine receptors whereas the bipolar cell terminals presynaptic to ONa RGCs express primarily GABA receptors. To address the role of inhibition in shaping ONa RGC output across light levels, we utilized the $GABA_Aa3$ receptor knockout mutant, which eliminates GABAergic and half of Glycinergic inhibition on ONa RGCs as well as the GABAergic presynaptic inhibition at bipolar cell terminals. We performed single cell patch-clamp recordings to measure light-evoked excitatory and inhibitory inputs and action potential output from ONa RGCs in mutant and control retina across a range of light levels. At dim light levels, the mutant ONa RGCs demonstrate a reduction in inhibitory currents consistent with a loss of inhibitory receptors, however, at bright light levels the magnitude of ONa inhibitory currents is similar between WT and mutant retina. Furthermore, at dim light levels, mutant ONa RGCs show a reduction in excitatory current amplitude but show a significant increase in excitatory current amplitude at bright light levels. Despite the decrease in synaptic excitation and inhibition under dim light conditions, we found that mutant ONa RGCs exhibit no change in their spike output compared to control cells at this light level, indicating a compensatory mechanism that operates by rescaling the excitatory and inhibitory inputs. On the contrary, mutant ONa RGCs exhibit an increase in the light-evoked spike output and a heightened sensitivity to contrasts at bright light levels, which could be the result of an increase in amplitude of the excitatory synaptic input. Together, our results demonstrate how alteration in inhibition differentially impacts the output of a key retinal circuit under dim vs bright light conditions and also how the ONa retinal circuit functionally compensates for loss of inhibition under dim light conditions.

Disclosures: A. Sawant: None. **R. Sinha:** None. **M. Hoon:** None.

Poster

PSTR146. Photoreceptors and Retinal Circuitry

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR146.10/Y5

Topic: D.06. Vision

Title: Rgc mirna-target networks driving visual system development

Authors: *A. CARVELLI[1], M. GHANEM[3], F. ZAMPA[2], N. ZOLBOOT[4], J. X. DU[5], G. LIPPI[4];
[2]The Scripps Res. Inst., [1]Scripps Res. Inst., San Diego, CA; [4]The Scripps Res. Inst., [3]The Scripps Res. Inst., La Jolla, CA; [5]UCSD, San Diego, CA

Abstract: The Retinogeniculate Tract (RT) is the first relay of the visual system, connecting the retina to the dorsal lateral geniculate nucleus (dLGN). The RT refinement requires the establishment of spatiotemporal signaling pathways. After birth, each dLGN neuron receives equal inputs from overlapping ipsi- and contralateral Retinal Ganglion Cell (RGC) axons. Through eye-specific segregation, within P2 and P10, each dLGN neuron ends up making synapses only with ipsi- or contralateral RGC axons. However, very little is known about the role of miRNAs in RT refinement. I took advantage of a short TNRC6B-derived peptide (T6B) able to bind AGO2, outcompeting with endogenous TNRC6, hence rapidly blocking global miRNA function. The selective miRNA LOF in RGCs was achieved by combining a floxed T6B version with the Cre protein driven by a Pan-RGCs specific promoter. To label ipsi- and contralateral RGC axons, floxed mRuby and iRFP proteins were used. Also, a mutant version of T6B (T6Bmut), a peptide not able to bind AGO2, was employed as a control. Intravitreal adenoviral injections of the aforementioned constructs were performed in P2 pups. Analysis of P10 flat-mount retinas confirmed the construct's expression in the RGCs. Further, P10 optic nerve analysis revealed well-organized RGC axons projecting to the dLGN both in T6Bmut and in the not injected condition. On the other hand, in the miRNA LOF condition, outcoming RGC axons were barely detectable. Strikingly, fluorescence analysis of ipsi- and contralateral RGC axons on P10 brain slices, revealed an impaired segregation on the dLGN surface in miRNA LOF condition. Both T6Bmut and the not injected condition did not show any segregation defects. Further, looking downstream in the RGC axons route toward the Superior Colliculus (SC), axonal elongation defects were highlighted. In particular, a reduced thickness in the axons bundle and a weaker mRuby/iRFP signal on the SC, in miRNA LOF condition, was detected. Taken together, these data suggest the involvement of miRNA in the visual system development and refinement. However, pinpointing the precise miRNA molecular mechanisms remains challenging. Each miRNA represses modestly and not uniformly hundreds of different targets, making it hard to understand the cascade of molecular events underlying LOF phenotypes. To map cell type-specific miRNA-target interactions in RGCs in vivo, I am using a novel enhanced Cross-Linking and ImmunoPrecipitation (eCLIP) technology, crossing Chrnb3-Cre line (RGC-specific) with tAGO mice. My goal is to map the RGC-specific miRNA-target networks orchestrating the RT development, puzzling out the tight interplay between RGCs and LGN neurons.

Disclosures: **A. carvelli:** A. Employment/Salary (full or part-time); The Scripps Research Institute. **M. Ghanem:** A. Employment/Salary (full or part-time); The Scripps Research Institute. **F. Zampa:** A. Employment/Salary (full or part-time); The Scripps Research Institute. **N. Zolboot:** A. Employment/Salary (full or part-time); The Scripps Research Institute. **J.X. Du:** A. Employment/Salary (full or part-time); The Scripps Research Institute. **G. Lippi:** A. Employment/Salary (full or part-time); The Scripps Research Institute.

Poster

PSTR146. Photoreceptors and Retinal Circuitry

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR146.11/Y6

Topic: D.06. Vision

Support: MEXT Grant S1101013
Senshu University

Title: The priming effect in the retina

Authors: *H. ISHIKANE;
Dept. of Psychology, Senshu Univ., Kanagawa, Japan

Abstract: Looming objects elicit protective behaviors in animals. For example, mice and frogs escape from looming objects. The neural basis of escape behavior is being elucidated, but the complete picture is not yet clear. The frog retina transmits highly abstracted visual information to the frog's brain than other animals. In addition, it has been demonstrated that class-4 neurons (a subtype of retinal ganglion cells called dimming detectors) transmit escape-related information to the frog's brain. In this study, we investigated the priming effect on escape behavior in frogs. To elucidate the neural basis of the priming effect, spikes were recorded from retinal ganglion cells in frogs. [Behavioral experiment] Two stimuli were presented successively to the bullfrogs (*Rana catesbeiana*), and their escape behavior was monitored. We investigated how the first stimulus, which cannot trigger the escape behavior by itself, affected the escape behavior triggered by the second stimulus. First, we confirmed that frogs showed escape behavior in response to a large expanding dark spot (maximum diameter: 35 degrees). Next, we presented a small expanding dark spot (maximum diameter: 8.75 degrees), which cannot elicit escape behavior by itself, and then presented a large expanding dark spot. The probability of triggering escape behavior was significantly higher than that in the control condition (no preceding small spot). These results suggest that the priming effect exists in visually evoked escape behavior in frogs. [Electrophysiology] The frog retina was isolated, and spikes were recorded from retinal ganglion cells using a multielectrode array. We focused on class-4 neurons which contribute to visually elicited escape behavior in frogs. A small expanding dark spot was presented onto the retina. No response was observed because the small spot was outside the receptive field of the recorded class-4 neuron. Then a large expanding dark spot was presented. Responses were observed because the receptive field of the recorded class-4 neuron was positioned inside the large spot. The number of spikes was significantly larger than that of spikes recorded in response to a large expanding dark spot without the priming stimulus (a small expanding spot). These results suggest that the priming effect observed in the visually elicited behavior of frogs could be explained by the output of the retina.

Disclosures: H. Ishikane: None.

Poster

PSTR146. Photoreceptors and Retinal Circuitry

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR146.12/Y7

Topic: D.06. Vision

Title: Reverse correlation using spatially correlated stimuli

Authors: *J. UMEMOTO, H. ISHIKANE;
Senshu Univ., Kawasaki-shi, Japan

Abstract: The reverse correlation is the systematic method for estimating receptive fields of visual neurons. Random noise stimuli, such as random checker patterns, are commonly used for estimating receptive fields of retinal ganglion cells. However, such stimuli have distinct characteristics compared to natural images: random noise stimuli contain little spatial correlation because the values of neighboring pixels are independent of each other and have a flat frequency spectrum exhibiting uniform energy distribution. In the present study, we propose to apply spatially correlated stimuli in reverse correlation. Correlated stimuli were generated by the following procedure. Initially, images of random checker patterns were each low-pass filtered using spatial filters of different sizes.Then, the filtered images were binarized. Conventional binary random checker stimuli were also prepared as control stimuli. Then we compared the spatial receptive field profile obtained by using correlated stimuli with that using random stimuli. The result showed that the spatial receptive field region was estimated to be wider than that obtained by using random noise stimuli. A possible reason is the continuity of stimulus. A correlated stimulus set contains images composed of low frequency roughly on the order of receptive field sizes, such stimuli are not spatially discretized, and they could have the potential to efficiently transmit signals from distal dendrites and neighboring cells to the soma of the recording cell. Additional validation was also performed to evaluate the efficiency in estimating the receptive field over time. We compared the estimated receptive fields in accordance with an increase in repeated trials of stimulus presentation. The spatiotemporal receptive fields were more separable from noise level with fewer trials compared to those obtained using random stimuli. These results suggest that adopting spatially correlated stimuli might be efficient in terms of detection ability to reverse correlation methods for estimating the receptive fields of visual neurons.

Disclosures: **J. Umemoto:** None. **H. Ishikane:** None.

Poster

PSTR146. Photoreceptors and Retinal Circuitry

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR146.13/Y8

Topic: D.06. Vision

Support: DFG (German Research Foundation) SFB 1456, Project-ID 432680300

Title: Leveraging convolutional neural networks to uncover computations in the inner retina

Authors: *M. VYSTRCILOVÁ[1], S. SRIDHAR[2], M. F. BURG[1], T. GOLLISCH[2], A. ECKER[1]; [1]Inst. of Computer Sci. and Campus Inst. Data Sci., Univ. of Göttingen, Göttingen, Germany; [2]Dept. of Ophthalmology, Univ. Med. Ctr. Göttingen, Göttingen, Germany

Abstract: Visual perception starts in the retina. This thin layer at the back of the eye transforms incoming light into electrical signals, laying the foundation for visual processing. Understanding the complex non-linear encoding mechanisms of the retina is fundamental to understanding vision. However, directly observing the intermediate processing stages of this neural network is challenging, and not possible at scale. Our goal in this project is to infer hidden computations in the retina, specifically those by wide-field amacrine cells, from the activity of large populations of ganglion cells. Here we employ a data-driven approach using a space-time-convolutional neural networks (CNNs). We present hours of natural and white-noise stimuli to marmoset and salamander retinas, while simultaneously recording the spiking activity of retinal ganglion cells (RGCs) using multi-electrode arrays. Leveraging these recordings, we train CNN models to predict the spiking activity of RGCs in response to the same stimuli. As our data-driven models have high predictive performance, we treat them as an in-silico digital twin of the retina. Applying interpretability techniques from machine learning to these digital twins enables us to gain insights into retinal processing not only at the input and output layers but also in the intermediate layers. As a first step, we leverage the CNNs to generate synthetic stimuli that maximize the predicted response of RGCs. Such stimuli allow us to unravel the visual features which RGCs are most sensitive to and compare processing of white noise and natural movie stimuli. In the future, we plan to combine such stimulus synthesis approaches with biological constraints on the network architecture to gain insights into the function of other cells in the intermediate layers of the retina, such as wide-field amacrine cells.

Disclosures: **M. Vystrcilová:** A. Employment/Salary (full or part-time); University of Göttingen. **S. Sridhar:** A. Employment/Salary (full or part-time); University Medical Center Göttingen. **M.F. Burg:** A. Employment/Salary (full or part-time); University of Goettingen. **T. Gollisch:** A. Employment/Salary (full or part-time); University Medical Center Göttingen. **A. Ecker:** A. Employment/Salary (full or part-time); University of Göttingen.

Poster

PSTR146. Photoreceptors and Retinal Circuitry

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR146.14

Topic: D.06. Vision

Support: A03-4451

Title: Flrt2-unc5-lphn signaling drives rejection of inappropriate synaptic partners in the retinal direction-selective circuit

Authors: *M. DEMBLA, C. PRIGGE, J. KAY;
Duke university, Durham, NC

Abstract: Precise and selective wiring of neuronal cell types is distinct feature of nervous system. The accuracy at which circuits are wired is important because it ensures that circuits are composed of cell types appropriately suited for its proper functioning. Proper wiring requires a complex molecular system of cell-cell recognition among developing neurons, so that inappropriate partners can be avoided while contacts with appropriate partners can be converted into synapses. While much is known about adhesion molecules that bring circuit partners together, cell-cell recognition mechanisms that mediate rejection of inappropriate partners remain unknown. Here we use the direction-selective (DS) circuit of mouse retina as a model system to investigate this issue. We show that the key DS circuit cell types, presynaptic starburst amacrine cells and postsynaptic ON-OFF direction-selective ganglion cells (ooDSGCs), avoid inappropriate partners using the FLRTs-LPHNs-UNC5 receptor-ligand system. FLRT2 is expressed by DS circuit neurons, which project arbors into two narrow sublayers of the inner plexiform layer (IPL) neuropil. UNC5C and UNC5D, meanwhile, localize to surrounding IPL sublayers. We find that growing DS-circuit dendrites sprout into adjacent $UNC5^{+}$ territories as a normal part of their dendritic development, and that growth of these mistargeted branches is suppressed via direct FLRT2-UNC5 binding. Genetic deletion of FLRT2 or UNC5s, or interference with their binding allows mistargeted arbors to expand in ectopic IPL sublayers, where they acquire inappropriate synapses from non-DS partners. Using a combination of in vitro synapse formation assays and in vivo misexpression, we show that UNC5s exert their inhibitory effects by interfering with FLRT2-LPHN3 adhesion and synaptogenesis. Correctly targeted nascent dendrites are selected for synapse formation and further growth due to strong FLRT2-LPHN adhesion, whereas mistargeted branches are eliminated due to addition of UNC5s to the FLRT2-LPHN3 complex which occludes synaptogenesis. Our results therefore define a molecular mechanism by which DS circuit neurons distinguish correct synaptic partners from inappropriate ones. FLRT, UNC5, and LPHN family molecules are widely expressed throughout the developing nervous system, so the mechanism we describe may be broadly relevant to development of synaptic specificity

Disclosures: **M. Dembla:** None. **C. Prigge:** None. **J. Kay:** None.

Poster

PSTR146. Photoreceptors and Retinal Circuitry

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR146.15/Y9

Topic: D.06. Vision

Support: 2020R1C1C101024514
2022M3E5E801739512

Title: Expression Pattern and eletrophysiological recordings of HCN2 Channel in Retina

Authors: *J. CHU, B. KANG, K. KIM, G.-S. HONG;
Korea Inst. of Sci. and Technol., Seoul, Korea, Republic of

Abstract: The HCN (Hyperpolarization-activated Cyclic Nucleotide-gated) channel is a voltage-gated cation channel expressed in various regions of the central nervous system and the retina. HCN2, one of the 4 subtypes of the HCN channels, is known to be primarily expressed in retina bipolar cells, which support transmission of visual signals from photoreceptors to ganglion cells. While HCN2 expression changes in retinal degeneration models have been described, the exact function and specific expression pattern have not yet been fully elucidated. Fluorescence In situ Hybridization (FISH) results found HCN2 channels highly expressed not only in ON bipolar cells but also in other cells (amacrine or interplexiform cell) present in the inner nuclear layer (INL). Patch clamp experiments were also conducted to observe the hyperpolarization-activated currents in specific cell types present in the INL. These results indicate that the HCN2, which can play a role in channel function, is expressed not only in ON bipolar cells but also in other cells and serves to modulate the retinal circuit.

Disclosures: J. Chu: B. Contracted Research/Research Grant (principal investigator for a drug study, collaborator or consultant and pending and current grants). If you are a PI for a drug study, report that research relationship even if those funds come to an institution.; 2020R1C1C101024514, 2022M3E5E801739512. **B. Kang:** B. Contracted Research/Research Grant (principal investigator for a drug study, collaborator or consultant and pending and current grants). If you are a PI for a drug study, report that research relationship even if those funds come to an institution.; 2020R1C1C101024514, 2022M3E5E801739512. **K. Kim:** B. Contracted Research/Research Grant (principal investigator for a drug study, collaborator or consultant and pending and current grants). If you are a PI for a drug study, report that research relationship even if those funds come to an institution.; 2020R1C1C101024514, 2022M3E5E801739512. **G. Hong:** B. Contracted Research/Research Grant (principal investigator for a drug study, collaborator or consultant and pending and current grants). If you are a PI for a drug study, report that research relationship even if those funds come to an institution.; 2020R1C1C101024514, 2022M3E5E801739512.

Poster

PSTR146. Photoreceptors and Retinal Circuitry

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR146.16/Y10

Topic: D.06. Vision

Support: Fonds de Recherche de Quebec-Sante # 301004
CONACYT #664900
BRAIN CANADA FOUNDATION-AZRIELI FOUNDATION

Title: Studying light-evoked retinal responses following optogenetic vision therapy

Authors: *K. D. ROJAS GARCÍA, N. ARNOLD, A. VILLEMAIN, S. TRENHOLM;
Integrated Program in Neurosci., Montreal Neurolog. Inst. and Hosp., Montreal, QC, Canada

Abstract: Background: Retinal degenerative diseases are a leading cause of vision loss and rise from the degeneration of rod and cone photoreceptors. Following photoreceptor degeneration, other cells in the retina remain intact, meaning that they could be targeted with optogenetics as a possible therapeutic strategy. However, the healthy retina sends out many distinct channels of visual information, via different functional types of retinal ganglion cells (RGCs), that each code different visual features, such as motion, contrast, image size, etc. Due to technical limitations, it may not be possible to restore all of these channels with optogenetic approaches. Our goal is to systematically assess how many functional retinal channels are restored when optogenetic tools are targeted to different retinal cell types, and what the implications are for vision if only a single channel of retinal information is restored.
Methods: Experiments are performed in rd1 mice, a model of retinitis pigmentosa, who lose vision due to photoreceptor degeneration by around P30. To express optogenetic tools in retinal neurons, we perform intravitreal injections with AAVs to express MW-Opsin in different retinal neurons via the use of cell-type-specific promoters. To test whether vision has been effectively restored, we screen injected mice with a light-room/dark-room test (a two-chamber arena in which sighted mice show a preference for the dark room). We also check the efficacy of injections using an in vivo retina epifluorescent microscope. Finally, to examine light responses of retinal ganglion cells, we place retinae on a 256-channel multi-electrode array and present the retina with a series of movies.
Results and Conclusions: We have successfully expressed optogenetic tools in the retina of blind animals and found that their vision is restored (at least as assessed with our light-room/dark-room screen). We have acquired light responses from wild type retinal ganglion cells and sorted them into functional cell types. We have obtained restored light responses from optogenetically-treated rd1 retinae and found these responses are predominantly of a single ON-type. To date, these results indicate successful restoration of a single retinal channel of information by targeting expression of a single type of opsin to a range of RGC types.

Disclosures: K.D. Rojas García: None. **N. Arnold:** None. **A. Villemain:** None. **S. Trenholm:** None.

Poster

PSTR146. Photoreceptors and Retinal Circuitry

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR146.17/Y12

Topic: D.06. Vision

Support: NIH Grant EY032095A

Title: Investigating the role of mechanotransduction in the accessory optic system

Authors: N. HAMILTON, PhD[1], K. CHAUDHARI[1], ***V. NECKLES**[2], J. KIRALY[3], A. KHERADMAND[4], A. KOLODKIN[1];
[2]Neurosci., [1]Johns Hopkins Univ., Baltimore, MD; [3]Johns Hopkins Med. Institutions, Baltimore, MD; [4]JOHNS HOPKINS UNIVERSITY, Baltimore, MD

Abstract: The optokinetic reflex (OKR) is an evolutionarily conserved pathway responsible for stabilizing images on the retina. OKR consists of slow-movement during object tracking followed by fast-movement that returns the eye to the initial position. The slow motion seen during OKR is directed by the accessory optic system (AOS). On direction selective ganglion cells (oDSGCs) of the AOS are tuned to various directions and show direction selectivity in four preferred directions - up, down, forward and backward. We generated an RNA sequencing dataset using Hoxd10-GFP mice, in which all AOS DSGCs are labeled, that has allowed for the identification of direction-specific DSGC markers. Interestingly, we find that putative forward oDSGCs show a selective enrichment in the expression of the mechanosensitive ion channel Piezo2. We detect Piezo2 expression in both monostratified and bistratified subsets of retinal ganglion cells (RGCs) which project to the NOT and DTN, areas of the brain, consistent with horizontally tuned AOS oDSGC targets. Loss of Piezo2 function in developing retina cells significantly impacts the horizontal OKR seen during object tracking, which is mediated by forward DSGCs. In these mice, the eye tracks horizontal movement in a diagonal direction, demonstrating a cross-coupling phenotype of image stabilization in the vertical and horizontal axes. Parallel assessments of human DA5 syndrome patients, who harbor gain-of-function *Piezo2* mutations, reveal an inability of the eye to move in an upward direction. We are currently focused on uncovering the importance of the Piezo2 mechanochannel in the development and function of forward-tuned oDSGCs.

Disclosures: **N. Hamilton:** None. **K. Chaudhari:** None. **V. Neckles:** A. Employment/Salary (full or part-time); Johns Hopkins University School of Medicine. **J. Kiraly:** None. **A. Kheradmand:** None. **A. Kolodkin:** None.

Poster

PSTR146. Photoreceptors and Retinal Circuitry

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR146.18/Y13

Topic: D.06. Vision

Support: NIH R01 EY029323
NIH R01 EY014454
NIH DP2 MH125812

Title: Optimizing Expansion Microscopy (ExM) for the mouse retina

Authors: *C. HERNANDEZ[1], J. A. MINEHART[2], C. ZHANG[3], C. M. SPEER[4];
[2]Neurosci., [3]Biophysics, [4]Biol., [1]Univ. of Maryland, Col. Park, College Park, MD

Abstract: The mouse retina contains a wide array of cell types that form parallel circuits for visual processing. The function of each circuit is determined in part by its specific pattern of synaptic connections as well as by the molecular organization of each synapse within the circuit. To complement electron microscopy-based reconstructions that define circuit patterns, studies of retinal circuits will benefit from the application of super-resolution optical imaging tools to reveal molecular information at specific synapses. Expansion Microscopy (ExM) is one super-resolution approach that achieves sub-diffraction-limit resolution using conventional confocal instruments, thereby opening new opportunities for broader adoption by researchers studying the retina. ExM involves the cross-linkage of biomolecules in tissue to a swellable hydrogel matrix, which is then expanded in water to physically separate and thereby resolve the positions of nearby targets. A number of different labeling strategies and hydrogel chemistries have been tested to improve ExM in brain tissue, but these have not been optimized for application to the retina. To help in this effort, we are developing an optimized ExM protocol for studying synaptic connections in the mouse inner retina. We have focused on imaging genetically-targeted retinal ganglion cells and inhibitory synaptic connections made by amacrine cells. Here we present results from our tests of pre- versus post-expansion immunostaining, distinct hydrogel and cross-linkage chemistries, and different tissue digestion protocols each with varying effects on label retention and expansion factor. Our results demonstrate that the molecular composition of retinal tissue is distinct from that of brain tissue, with impacts on expansion factor and image resolution when using brain-optimized protocols. We are developing our retina-specific protocol as an open-access resource for application and improvement by the retina and ExM research communities.

Disclosures: C. Hernandez: None. **J.A. Minehart:** None. **C. Zhang:** None. **C.M. Speer:** None.

Poster

PSTR146. Photoreceptors and Retinal Circuitry

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR146.19/Y14

Topic: D.06. Vision

Support: NIH Grant EY033551

Title: Caspase-dependent CAM and BIGH3-Induced HRP Apoptosis

Authors: *A. TSIN[1], L. VALDEZ[2], L. GUERRA[2], A. RODRIGUEZ[2];
[1]Neurosci., Univ. of Texas Rio Grande Valley, Edinburg, TX; [2]Neurosci., Univ. of Texas Rio Grande Valley Sch. of Med., Edinburg, TX

Abstract: Human retinal pericytes (HRP) are contractile cells that provide support for endothelial cells of capillaries, which are essential in the regulation of retinal vasculature in the eye. Early stages of diabetic retinopathy (DR) are characterized by the loss of retinal pericytes, which lead to the development of advanced-stage pathology including angiogenesis. Although much is known about the etiology of DR, the apoptotic pathway that incites retinal pericyte loss remains unclear. It is known that α3β1 integrin plays a role in the induction of apoptosis in pericytes through angiopoietin and caspases 3, 8, 10 and 14 have been reported to be involved in pericyte apoptosis. TGFβ-Induced Gene Human Clone 3 (BIGH3) is a downstream target molecule of TGFβ, which is known to contribute to HRP apoptosis under hyperglycemic conditions. While integrins have an important role in cell maintainability with integrin-mediated cell and ECM activity, perturbing the interaction can result in a caspase-initiated apoptosis, such as integrin mediated death (IMD). In this present study, HRP was used to determine the apoptotic response to Camptothecin (CAM) and BIGH3. HRP were grown with 10% FBS in Complete Classic Media in a humidified 5% CO_2 incubator at 37°C. Early passages of HRP (passages 5-7) were seeded on a 24 well plate at $5x10^4$ cells per well and passaged on five P100 dishes. Once cells were confluent, they were then treated with 3mM of CAM or 5ug/mL of BIGH3 for 48 and 72 hours. Cleaved Caspase 3 expression was measured by Western Blot analysis and apoptosis was identified using an AO/PI stain. Treatment with CAM and BIGH3 resulted in a significant increase in cell death compared to negative controls. Moreover, % cell apoptosis also increased with time. Immunoblotting shows an expression of Cleaved Caspase 3 in cells treated with CAM and BIGH3 (but not in vehicle-only controls), in which cleaved caspase protein increased from 48 to 72 hours. Therefore, these results strongly suggest that CAM and BIGH3 induced HRP apoptosis is Caspase-Dependent.

Disclosures: **A. Tsin:** None. **L. Valdez:** None. **L. Guerra:** None. **A. Rodriguez:** None.

Poster

PSTR146. Photoreceptors and Retinal Circuitry

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR146.20/Y15

Topic: D.06. Vision

Support: DP2 EY027983
R21 EY030565
Brain Research Foundation Fay/Frank Seed Grant

Title: Genetic underpinnings of ipRGC diversity

Authors: M. ARANDA, O. PAYAN PARRA, T. YAMADA, Y. YANG, ***T. SCHMIDT**; Northwestern Univ., Evanston, IL

Abstract: Light is a profoundly important regulator of physiology and behavior across a wide range of organisms. Light information is relayed via diverse retinal ganglion cell types to

approximately 50 distinct targets in the brain. The melanopsin-expressing, intrinsically photosensitive retinal ganglion cells (ipRGCs) represent 6 of the ~40-50 retinal ganglion cell types present in the mouse retina. M1-M6 ipRGCs are defined by a distinct complement of subtype-defining morphological, physiological, and transcriptional characteristics. However, how this cellular diversity is achieved is largely unknown. Brn3b (Pouf42) is a transcription factor involved that is poised to influence gene expression programs defining properties of ipRGCs. In this study we tested the hypothesis that Brn3b actively shapes the morphological, physiological, and transcriptional identity of ipRGC subtypes. We compared gene expression patterns, melanopsin expression, morphological properties as well as ipRGC-driven behaviors in mice where Brn3b is conditionally removed (Brn3bcKO animals) or overexpressed (Brn3bOE) in ipRGCs. Our results indicate that Brn3b expression levels in ipRGC correlates with, and actively regulates, the levels of melanopsin mRNA and protein. Using TRAP-seq we found that Brn3b is a critical regulator of transcriptional programs that define ipRGC subtype identity. Additionally, we found that Brn3b plays a key role defining morphological properties such as soma size and dendritic development of ipRGC subtypes. Finally, using intersectional virus approaches we analyzed the axonal projections patterns to the main ipRGC targets in the brain as well as ipRGC-driven behaviors. We found that ipRGC-projection patterns as well as ipRGC-driven behaviors were altered in Brn3bcKO and Brn3bOE mice. Altogether these findings indicate that Brn3b define the transcriptional identity, the morphological properties and behaviors driven by ipRGC subtypes.

Disclosures: **M. Aranda:** None. **O. Payan Parra:** None. **T. Yamada:** None. **Y. Yang:** None. **T. Schmidt:** None.

Poster

PSTR146. Photoreceptors and Retinal Circuitry

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR146.21/Y16

Topic: D.06. Vision

Support: NIH DP2 EY022584
NIH T32 EY025202

Title: Whole brain atlas of light induced activity in the mouse

Authors: *J. D. BHOI, A. WCISLAK, T. M. SCHMIDT;
Neurobio., Northwestern Univ., Evanston, IL

Abstract: Light is one of the most fundamental features of an animal’s environment and modulates behavior and physiology to allow animals to adapt to changes in their environment. Retinal ganglion cells (RGCs), the output cells of the retina, are a diverse class of over forty cell types, each of which sends highly processed visual information to more than 50 retinorecipient targets in the brain. The intrinsically photosensitive (ip)RGCs are a class of RGCs which express

the photopigment melanopsin, endowing them with sensitivity to light independent from rod and cone input. ipRGCs project to more than 20 of the 50 retinorecipient brain regions in the mouse and play an essential role in both subconscious visual functions and conscious visual perception. While the RGC projections to the brain have been extensively mapped, the full extent of how light input from the retina functionally impacts the brain through RGCs, and how ipRGCs specifically contribute to this activation, remains an open question. The goal of the current study is to create a whole brain map of light-induced cFos expression and to identify the brain areas where cFos expression is driven by ipRGCs.
To stimulate RGCs, we exposed both male and female mice to a 15-minute pulse of 1000 lux, broad spectrum light. Following light exposure, we used immunohistochemistry to stain brain sections for the immediate early gene cFos. We then registered individual brain sections to the Allen Mouse Brain Common Coordinate Framework and quantified cFos expression. Further, to test the if melanopsin phototransduction is necessary for light induced cFos, we repeated these experiments in melanopsin knockout mice. If a brain region is activated by light, then we should detect cFos expression following light stimulation. Likewise, if that activation is mediated by ipRGCs and melanopsin, then we expect to see an attenuation of cFos expression in melanopsin knockout mice compared to controls. We find that light induces cFos expression in many brain regions, including canonical visual regions such as the lateral geniculate nucleus and non-canonical visual regions such as the medial amygdala. Further, we observed cFos immunoreactivity in brain regions which do not receive direct retinal input, indicating that this technique can reveal regions downstream of retinorecipient areas. This atlas contributes to our current understanding of visual processing, and it will serve as a critical resource for research investigating the influence of light on a wide range of neural circuits.

Disclosures: **J.D. Bhoi:** None. **A. Wcislak:** None. **T.M. Schmidt:** None.

Poster

PSTR146. Photoreceptors and Retinal Circuitry

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR146.22/Y17

Topic: D.06. Vision

Support: NIH grant EY031411

Title: Retinal circuit contributions to luminance adaptation in primate foveal and peripheral retina

Authors: *A. H. MILLER, A. SAHA, R. SINHA;
Neurosci., Univ. of Wisconsin-Madison, Madison, WI

Abstract: We experience a range of light intensities over the course of a day that far exceeds the dynamic range of neuronal signaling in the retina and yet our visual perception of object brightness remains constant. Light adaptation ensures brightness constancy by adjusting the gain

of neuronal responses as illumination varies. Most of what we know about light adaptation in the primate retina comes from studies in the peripheral retina which have identified two distinct sites of adaptation—one at cone photoreceptors which operates at brighter light levels and the other at downstream circuitry which drives light adaptation in the retina at lower light levels and relies on pooling of cone inputs. However, we know nothing about the relative contribution of photoreceptor vs circuit mechanisms towards light adaptation in the primate fovea—the specialized region in the central most part of the primate retina responsible for our high-acuity vision. In the fovea, the dominant neural circuit - midget ganglion cells - receive signals originating from a single cone photoreceptor unlike in the peripheral retina where retinal circuits pool input from a population of cones. To test if light adaptation is different in the primate fovea, we used single-cell electrophysiology to measure light-evoked electrical responses from primate foveal midget ganglion cells as well as cone photoreceptors across a range of background luminance. We compared adaptation in both the fovea and periphery. Unlike in the peripheral retina, recordings in the fovea show that light adaptation measured at the level of foveal midget ganglion cells is identical to that in cone photoreceptors across all light levels. There is no circuit adaptation at lower background luminance typically observed in peripheral ganglion cells including midget ganglion cells. These results indicate that the mechanisms for luminance adaptation in primate foveal versus peripheral retina differ and these differences correlate with changes in retinal circuit convergence.

Disclosures: **A.H. Miller:** None. **A. Saha:** None. **R. Sinha:** None.

Poster

PSTR146. Photoreceptors and Retinal Circuitry

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR146.23/Y18

Topic: D.06. Vision

Support: NIH Grant EY029772
That All May See Foundation

Title: Severity of cone ablation differentially affects tuning properties of ON-direction selective ganglion cells

Authors: *A. BALRAJ, S. HARRIS, F. A. DUNN;
Univ. of California San Francisco, San Francisco, CA

Abstract: Visual acuity can remain unaffected after dysfunction of up to half of all cones, suggesting insensitive diagnostics and/or compensatory mechanisms within the visual system. Previous work from our lab has shown that compensation can occur in ON-sustained alpha retinal ganglion cells (RGCs), which alter their spatiotemporal filtering in response to partial cone loss. However, the reaction of other RGC types to fewer cone inputs remains unknown. We aimed to examine the effects of cone loss on ON-direction selective ganglion cells (oDSGCs),

which are important to the accessory optic system rather than to conscious visual perception. Our lab has found differences in response gain and tuning properties of oDSGCs sensitive to upward- (Superior) and downard- (Inferior) motion. Using a mouse model of partial cone ablation and electrophysiological recordings, we tested the hypothesis that graded cone loss correlates with reduced response gain in oDSGCs from reduced cone inputs. To study cone loss in the mature mouse retina, we generated a transgenic line (cone-DTR) where the simian diphtheria toxin receptor (DTR) is driven under a promoter for the M-opsin gene. Injection of diphtheria toxin (after P30) selectively ablates cones expressing M-opsin, and the dosage of diphtheria toxin was adjusted to induce 50% or 25% cone loss. To selectively target Superior and Inferior oDSGCs in the retina, we injected a retrograde tracer into the projection site of oDSGCs in 50% cone-DTR, 25% cone-DTR, and wildtype (WT) retinas. Mice were dark-adapted overnight and retinas were harvested for electrophysiological assessment. We used cell-attached recordings of labeled RGCs in response to a slowly drifting bar moving in 8 directions to measure spike responses to all directions (tuning curve area) and direction selectivity indices (DSI, the vector sum divided by the scalar sum of responses) of oDSGCs. In 50% cone-DTR retinas, oDSGCs had smaller tuning curve areas and fewer spikes in their preferred directions. Superior and Inferior oDSGC populations had lower tuning curve areas, and the variance of preferred direction increased in Superior oDSGCs. Tuning curve area and preferred direction responses were not significantly different between oDSGCs in 25% cone-DTR and WT retinas. However, oDSGCs in both cone loss conditions had larger DSI than WT controls, indicating greater direction selectivity. Our data demonstrate that the severity of cone loss differentially affects tuning of oDSGCs, where minor cone loss increases direction selectivity and further disruption reduces response gain.

Disclosures: **A. Balraj:** None. **S. Harris:** None. **F.A. Dunn:** None.

Poster

PSTR146. Photoreceptors and Retinal Circuitry

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR146.24/Y19

Topic: D.06. Vision

Support: DP2 EY027983
Klingenstein-Simons Fellowship
Sloan Research Fellowship

Title: Visual circuits underlying threat anticipation

Authors: *M. ARANDA, E. MIN, L. LIU, A. SCHIPMA, T. M. SCHMIDT;
Northwestern Univ., Evanston, IL

Abstract: The ability to anticipate potential threats in nature provides a clear evolutionary advantage. In mammals, the visual system is one of the primary senses used to respond and to detect changes in the environment. However, whether and how the visual system may influence

the ability to anticipate environmental threats is unknown. In this work, we tested the hypothesis that the melanopsin (Opn4)-expressing, intrinsically photosensitive retinal ganglion cells (ipRGCs) are critical for the ability to anticipate the future appearance of a previously experienced threat. For this, we developed a novel Visual Threat Anticipation (VITA) paradigm, in which we exposed control and melanopsin knock-out ($Opn4^{-/-}$) mice to a threatening visual "looming" stimulus (Exposure Phase). After two days, we returned animals to the identical context and measured their anticipatory behavior (i.e. VITA) in the absence of any "looming" stimulus to determine whether they associated that context with the prior threat exposure (Test Phase). Surprisingly, we found that male, but not female, $Opn4^{-/-}$ animals lacked VITA, indicating that ipRGCs are required for this behavior. VITA was also decreased in animals whose ipRGC are not able to release glutamate (Opn4Cre/+ $Vglut2^{fl/fl}$), while we observed an increased VITA in animals lacking GABA release in ipRGCs (Opn4Cre/+ $Gad^{2fl/fl}$) suggesting that glutamatergic ipRGCs increase anticipatory behaviors while GABAergic ipRGCs dampen VITA. To identify candidate circuits for VITA, we used c-Fos induction during the Test Phase of our VITA paradigm, and identified the Perihabenular Region (PHb) as candidate ipRGC-central target for driving VITA. In agreement with PHb involvement, chemogenetic silencing of GABAergic PHb neurons abolishes VITA, and chemogenetic excitation of PHb-projecting ipRGCs in $Opn4^{-/-}$ mice rescues VITA behavior. Our results suggest that ipRGCs mediate VITA behavior through a retina-PHb circuit.

Disclosures: **M. Aranda:** None. **E. Min:** None. **L. Liu:** None. **A. Schipma:** None. **T.M. Schmidt:** None.

Poster

PSTR146. Photoreceptors and Retinal Circuitry

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR146.25/Y20

Topic: D.06. Vision

Support: NIH Grant EY026978
NIH Grant EY034001
NIH Grant EY023341
NIH Grant EY027411
NIH Grant EY030623
NIH Vision Core grant EY0268

Title: Encoding Strategies of Natural Movies by Morphologically Distinct Bipolar Cells in the Retina

Authors: *J. HSIANG[1,2], N. SHEN[2], F. SOTO[2], D. KERSCHENSTEINER[2,3,4,5];
[1]Ophthalmology & Visual Sci., [2]Dept. of Ophthalmology and Visual Sci., [3]Dept. of Neurosci., [4]Dept. of Biomed. Engin., [5]Hope Ctr. for Neurolog. Disorders, Washington Univ. Sch. of Med., St. Louis, MO

Abstract: Understanding the encoding of the visual world within the retina is crucial for gaining insights into the subsequent processing of visual information along visual pathways. Bipolar cells (BCs) play a significant role in this process by integrating signals from photoreceptors and activating amacrine and ganglion cells within the retina, which are responsible for encoding a variety of visual features. Previous research has demonstrated the presence of distinct responses of bipolar cells to visual stimuli; however, several critical questions remain unaddressed, including: (1) the functional separation of morphologically distinct types, (2) the manner in which naturalistic movies are encoded by bipolar cells, and (3) the potential for different axonal branches of a single BC to encode distinct visual features.
In the present study, we sought to address these questions by targeting individual BCs, recording calcium signals via two-photon microscopy, morphologically identifying different types, and utilizing data augmentation in the differential encoding space to shed light on the mechanisms responsible for encoding complex visual features. Our findings revealed that while morphological types exhibit varied responses, they do not separate uniformly. The encoding space of naturalistic movies is predominantly characterized by differences in surround strength and transience. Notably, heterogeneous responses within a single BC were not spatially organized and were distance-independent, largely stemming from intrinsic noise. This study advances our understanding of the functional organization of bipolar cells and their role in encoding visual information within the retina.

Disclosures: **J. Hsiang:** None. **N. Shen:** None. **F. Soto:** None. **D. Kerschensteiner:** None.

Poster

PSTR146. Photoreceptors and Retinal Circuitry

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR146.26/Z1

Topic: D.06. Vision

Support: Sinergia Grant CRSII5_173728
Eccellenza Grant PCEFP3_187001
ERC Advanced Grant 694829

Title: Retinal ganglion cells in the human retina synchronize action potential arrival times at the optic disc

Authors: *A. BUCCI[1], M. BUETTNER[1], N. DOMDEI[2], F. B. ROSSELLI[1], R. DIGGELMANN[3], M. ZNIDARIC[1], B. ROSKA[1], A. HIERLEMANN[3], W. M. HARMENING[2], F. FRANKE[1];
[1]Inst. of Mol. and Clin. Ophthalmology Basel (IOB), Basel, Switzerland; [2]Univ. of Bonn, Bonn, Germany; [3]Dept. of Biosystems Sci. and Engin., ETH Zürich, Basel, Switzerland

Abstract: Precise timing of action potentials is crucial for processing sensory information. Axonal length and propagating speed largely determine the time necessary for action potentials

to reach postsynaptic neurons. Within the retina, retinal ganglion cell (RGC) axons form the retinal nerve fiber layer (RNFL), a highly organized layer with species-specific axonal arrangements. The human RNFL is characterized by unmyelinated axons, resulting in slow signal transmission, and by the presence of the fovea, a specialized region enabling high-resolution vision, which is located temporal to the optic nerve head (i.e., optic disc). Its central part, known as fovea centralis, is mostly devoid of RGC somas and axons. As a result, foveal cones establish connections with RGCs that are radially displaced on a ring-like structure encircling the fovea centralis. Thus, RGC axons originating temporal to the fovea are significantly longer than axons originating nasally, which extend directly towards the optic disc. This leads to different paths for visual information from adjacent cones in the fovea centralis to reach the optic disc, either through direct routes or longer trajectories circumnavigating the fovea centralis. We investigated whether different axonal lengths in the human RNFL entail distinct action potential traveling speeds, thereby synchronizing visual information. To measure traveling speeds precisely, we recorded action potentials of foveal RGCs in ex vivo human retinal explants at subcellular resolution by means of high-density microelectrode-arrays (HD-MEAs). We found that action potential propagation speeds varied based on the location of RGC somas relative to the fovea centralis. Foveal RGC axons originating temporal to the fovea centralis exhibited up to 50% faster action potential propagation speeds than those originating nasally. Moreover, peripheral RGCs exhibited action potentials propagating up to three times faster than those in the foveal retina. Both foveal and peripheral retina showed a bimodal distribution of propagation speeds among the two predominant cell types in primate retina: midget and parasol cells. By developing a comprehensive model of the human RNFL, we successfully predicted the entire paths and lengths of RGC axons, which strongly correlated with observed lengths and propagation speeds. Our findings suggest a compensatory mechanism in the human retina that contributes to synchronizing the arrival times of visual signals in the brain.

Disclosures: **A. Bucci:** None. **M. Buettner:** None. **N. Domdei:** None. **F.B. Rosselli:** None. **R. Diggelmann:** None. **M. Znidaric:** None. **B. Roska:** None. **A. Hierlemann:** None. **W.M. Harmening:** None. **F. Franke:** None.

Poster

PSTR146. Photoreceptors and Retinal Circuitry

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR146.27/Z2

Topic: D.06. Vision

Support: The Intramural Research Programs of the National Eye Institute
Deutsche Forschungsgemeinschaft (DFG, German Research Foundation) SFB 1381 (project-ID 403222702)

Title: Brorin/vwc2 modulates the activity of AMPA receptors in the retina through interaction with their core subunits and soluble associated proteins

Authors: *S. TOMAREV[1], N. NAKAYA[1], A. HOOVER[1], J. SCHWENK[3], R. N. FARISS[2], B. FAKLER[4];
[1]Section of Retinal Ganglion Cell Biology, Lab. of Retinal Cell and Mol. Biol., [2]Imaging Core, Natl. Eye Institute, NIH, Bethesda, MD; [3]Univ. of Freiburg, Univ. of Freiburg, Freiburg, Germany; [4]Inst. of Physiol., Inst. of Physiol., Freiburg, Germany

Abstract: Brorin, also known as Vwc2, is a secreted protein component of AMPA-type glutamate receptors (AMPARs) in the brain. The functions of Vwc2 in the retina have not been characterized, and the objective of this study was to investigate these functions using mice as a model organism. The composition of Vwc2-containing AMPARs in the retina is different from those in the brain as demonstrated by quantitative proteomics. Several auxiliary proteins of AMPARs, such as TARP γ2, TARP γ7, CNIH2, CNIH3, GSG1L, and SHSA9, were immunoprecipitated from brain but not from retinal membrane-enriched fractions using Vwc2 antibodies. In the retina of adult mice, expression of the *Vwc2* gene was detected mainly in horizontal and amacrine cells, with lower levels of expression also detected in retinal ganglion cells (RGCs). The outer plexiform layer of *Vwc2* knockout (KO) retina was thinner than in wild type (WT) retina (11.8±0.3 µm vs 14.9±0.4 µm, $p<0.001$). In adult *Vwc2* KO, the number of amacrine and horizontal cells declined compared with WT by 27% and 39%, respectively, while the number of RGCs remained unaltered. The levels of GluA1-GluA4 core subunits did not change significantly, while the levels of Noelins 1- 3 (also known as Olfactomedins 1-3), extracellular constitutents of surface AMPARs, was reduced by 40-50% in AMPARs isolated from *Vwc2* KO with a mixture of antiGluA1-4 antibodies. In *Vwc2* KO samples, spontaneous excitatory postsynaptic currents of cultured amacrine cells *in vitro* and in isolated retina showed a reduction in their number, peak amplitude and displayed an extended decay time compared with WT samples. The ratio of AMPAR GluA2/GluA1 core subunits was decreased at the surface membrane of *Vwc2* KO amacrine cells in association with increased Ca^{2+} permeability within these same cells compared with WT. There was no change in intracellular Ca^{2+} within RGCs between *Vwc2* KO and WT. For better mechanistic understanding, we studied direct interactions of Vwc2 with several proteins of the AMPAR complexes in co-transfection experiments. We found that Vwc2 directly interacts with GluA2, and Noelin 2 facilitates this interaction. Altogether, our results identify Vwc2 as a new and potentially crucial player affecting AMPAR activity through interaction with the core subunits and soluble associated proteins of AMPARs in the retina.

Disclosures: S. Tomarev: None. **N. Nakaya:** None. **A. Hoover:** None. **J. Schwenk:** None. **R.N. Fariss:** None. **B. Fakler:** None.

Poster

PSTR146. Photoreceptors and Retinal Circuitry

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR146.28/Web Only

Topic: D.06. Vision

Support: United States-Israel Binational Science Foundation (BSF #2021289)
Israel Science Foundation (ISF #2389/22)

Title: Seeing in the dark: high-order visual functions under scotopic conditions

Authors: *D. C. ELUL[1,2], A. MCKYTON[2], N. LEVIN[2];
[1]Edmond and Lily Safra Ctr. for Brain Sci. (ELSC), Hebrew Univ. of Jerusalem, Jerusalem, Israel; [2]Dept. of Neurology, Hadassah Med. Organization and Fac. of Medicine, Hebrew Univ., Jerusalem, Israel

Abstract: In many cases, people are able to function visually in almost complete darkness. However, high-level vision has not been investigated thoroughly under scotopic conditions, where only rod photoreceptors are active. Under these conditions, acuity is low, and a foveal scotoma spreads for about 1 degree in the center of the visual field. We set out to investigate how these limitations affect performance and eye movements in foveal tasks such as reading and face recognition. We recorded eye movements while testing the speed and accuracy of reading and of upright and inverted face matching under photopic and scotopic conditions. We also tested scotopic acuity and crowding at different eccentricities. Under scotopic conditions, relative to photopic conditions, participants read slower but accurately and showed a similar face inversion effect. Surprisingly, despite the foveal scotoma, fixations in both tasks were executed to similar locations as under photopic conditions; however, the duration of fixations was longer. For reading, the lack of use of peripheral vision could be explained by the crowding experiment results, which showed that scotopic crowding, similarly to photopic crowding, increases with eccentricity. For face recognition, it might be explained by the unharmed holistic nature of face recognition, which uses large receptive fields to achieve global perception. These results suggest that high-level visual tasks, even those that rely on foveal input, are solved in a similar manner under scotopic and photopic conditions.

Disclosures: D.C. Elul: None. **A. McKyton:** None. **N. Levin:** None.

Poster

PSTR147. Visual Responses During Behavior I

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR147.01/Z3

Topic: D.06. Vision

Support: Max Planck Society

Title: Mixed selectivity in mouse V1 revealed with visual stimulation in freely moving virtual reality

Authors: *M. K. MCCANN, T. BONHOEFFER, M. HUEBENER, D. A. GUGGIANA NILO;
Synapses - Circuits - Plasticity, Max Planck Inst. for Biol. Intelligence, Martinsried, Germany

Abstract: Neurons in the primary visual cortex (V1) exhibit tuning to a variety of visual stimulus features, like luminance, direction and orientation, spatial and temporal frequency, and stimulus location in visual space. Recent experiments in awake, head-fixed animals have revealed that V1 neurons are also modulated, or even driven, by non-visual inputs such as locomotion, position, and feedback mismatch. Very recent results in freely moving rodents show that V1 neurons exhibit luminance-dependent responses to head orienting movements, and that the spatial layout of visual receptive fields (RFs) in mice is similar to that of RFs found in head-fixed animals. However, to date no other visual tuning properties have been examined in unrestrained contexts. To close this gap, we use 1-photon wireless miniscopes to record calcium transients while presenting drifting grating Gabor patches to freely moving mice in a virtual reality (VR) arena. This allows recording of neural activity during presentation of a stimulus fixed in the animal's field of view, while the mouse moves unrestrained throughout the arena. We image the same field of view under both freely moving and head fixed conditions, allowing for direct comparison between responses of the same neurons. We find that neurons also display direction and orientation selectivity under freely moving conditions, with similar proportions of orientation and direction selective cells as previously reported in head-fixed experiments (88% and 33%, respectively). However, we report that the direction and orientation tuning of individual V1 neurons are more variable under freely moving conditions. We find both subpopulations of tuned cells where the preferred orientation or direction changes between freely moving and head fixed conditions, and other subpopulations where cells maintain their selectivity. Overall, we present an experimental paradigm that allows for the precisely controlled presentation of visual stimuli to freely moving animals. Preliminary data show that V1 neurons continue to exhibit direction and orientation tuning under unrestrained conditions, while response variability between freely moving and head fixed conditions suggests that inputs driven by locomotion and head or body movements interact with representations of visual stimulus features.

Disclosures: M.K. McCann: None. **T. Bonhoeffer:** None. **M. Huebener:** None. **D.A. Guggiana Nilo:** None.

Poster

PSTR147. Visual Responses During Behavior I

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR147.02/Z4

Topic: D.06. Vision

Support: JSPS Reader
Research Foundation for Opto-Science and Technology

Title: A visual discrimination task in head-fixed mice to understand the integration of visual stimuli projecting to both brain hemispheres.

Authors: *S. NISHIMOTO, M. WATANABE, A. FUNAMIZU;
Univ. of Tokyo, Bunkyo-ku, Japan

Abstract: Our brain represents multiple visual features such as shape, color, and texture in different brain regions in both hemispheres. The brain integrates the visual components to form a unified visual perception, but the detailed neural circuit to solve the binding problem is unclear. As many studies performed experiments in the binocular visual field, it is particularly unclear how the visual stimuli of the right and left fields, which are independently represented in the contralateral hemisphere of the primary visual cortex, are integrated to form visual perception. Here we test how the brain combines visual inputs in both brain hemispheres to form a unified visual perception. We designed a visual discrimination task in head-fixed mice. Mice were required to compare the stimuli presented on left and right screens by integrating the visual information from their left and right hemisphere. In the task, we presented small square-shaped visual stimuli simultaneously on the left and right screens for 50 milliseconds. These stimuli had relative luminance values (0, 63, 255) and were positioned at approximately 26.6 degrees lateral from the midline of the mouse to allow only one eye can see the ipsilateral stimulus. If the left stimulus had higher luminance than the right, the mice received sucrose water by licking a spout (Go trial). In contrast, when the right screen displayed a higher luminance stimulus than the left, the mice had to refrain from licking for 1 second (No-Go trial). Failure to resist licking during the No-Go trial resulted in a prolonged inter-trial interval (3 seconds instead of 0.5 seconds) as a punishment. After a month of training, the mice achieved an accuracy of 92.1% and 95.9% during Go trials for 0 vs. 63 and 63 vs. 255 conditions, respectively. On the other hand, No-Go trials had an accuracy of 91.2% and 62.3% for 0 vs. 63 and 63 vs. 255, respectively. To investigate the neural mechanisms underlying visual integration, we plan to electrophysiologically record the neural activity of the primary visual cortex and the prefrontal cortex during the task.

Disclosures: S. Nishimoto: None. **M. Watanabe:** None. **A. Funamizu:** None.

Poster

PSTR147. Visual Responses During Behavior I

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR147.03/Z5

Topic: D.06. Vision

Support: NIH grant EY002874
"MPS is a recipient of the RPB Disney Award for Amblyopia Research"

Title: Exploring the role of VIP neurons in visual perceptual learning in mice

Authors: *A. BELHE[1], N. BHATLA[2], B. RAKELA[2], M. HOSEINI[2], S. BAZARINI[2], S. HOSSEINCHI GHAREHAGHAJI[2], M. STRYKER[2];

[1]Physiol., [2]Physiology, Kavli Inst. for Fundamental Neurosci., Univ. of California, San Francisco, San Francisco, CA

Abstract: Visual perceptual learning is characterized by an enhancement in visual ability through practice. It enables individuals to recognize fine features and patterns that would otherwise go unnoticed. This type of learning has been observed in humans and animals, including monkeys, and correlates with changes in neural activity in both the primary visual cortex (V1) and higher visual areas. While the importance of practice and attention in visual perceptual learning is well-established, the specific brain circuitry responsible for inducing this learning process remains largely unknown. Here we introduce a behavioral procedure to investigate mechanisms of perceptual learning in head-fixed mice using virtual reality. In a T-maze, we train mice to run toward a cylinder displaying horizontal stripes and to avoid a cylinder with stripes of a different orientation. Mice are initially trained to discriminate horizontal stripes from vertical stripes. This learning immediately generalizes such that mice can discriminate the horizontal orientation from a novel diagonal (45º) orientation rotated away from horizontal in either the clockwise (CW) or counterclockwise (CCW) directions. One hallmark of perceptual learning in humans is that the enhanced ability to discriminate stimuli after repeated exposure is specific to the exposed stimulus and does not generalize to similarly difficult but different stimuli. Therefore, to determine whether mice exhibit stimulus-specific learning, we further trained mice to discriminate horizontal stripes from stripes rotated by 15º CW. Strikingly, these trained mice fail to discriminate horizontal stripes from those rotated 15º CCW, though with additional training they can learn to discriminate. These results demonstrate that we have developed a new assay for testing the stimulus-specific component of perceptual learning in mice. To further explore the neural substrates underlying perceptual learning, we have focused on the role of the VIP neurons. Activity of VIP neurons enhances neural responses and plasticity in mouse V1, suggesting that it may be required for perceptual learning. To test this hypothesis, we selectively ablated VIP neurons in VIP-Cre mice by injecting a Cre-dependent caspase virus after CW training, with the expectation that mastering the fine CCW discrimination would be retarded. Control mice received artificial cerebrospinal fluid injections. The viral injections were effective in eliminating most VIP cells in V1. Initial data obtained from a small number of experimental animals have yielded variable effects and is therefore inconclusive. Additional experiments are in progress.

Disclosures: **A. Belhe:** None. **N. Bhatla:** None. **B. Rakela:** None. **M. Hoseini:** None. **S. Bazarini:** None. **S. Hosseinchi Gharehaghaji:** None. **M. Stryker:** None.

Poster

PSTR147. Visual Responses During Behavior I

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR147.04/Z6

Topic: D.06. Vision

Support: NIH R01EY030998
NIH R21EY025403
NIH R00EY032179

Title: Spatiotemporal convolutional neural networks of primary visual cortex during free-viewing of natural scenes

Authors: *D. GALOR[1], J. MITCHELL[2], D. A. BUTTS[3], J. L. YATES[1];
[1]Univ. of California, Berkeley, Berkeley, CA; [2]Univ. of Rochester, Pittsford, NY; [3]Univ. of Maryalnd, College Park, MD

Abstract: Convolutional neural networks (CNNs) have emerged as a benchmark model of visual cortical areas. Spatiotemporal CNNs fit directly to neural data have been shown to capture a range of nonlinear response properties in salamander retina and V1 of mice, but in modeling primate visual cortex, CNNs have largely been spatial only, generating spike counts to static images. Here we sought to characterize how well data-driven CNNs capture responses at high time resolution (~5ms) generated during free-viewing of natural scenes. We focused our interrogation on how well models generalize to stimulus conditions that were not in the training set and how well models capture the transient response dynamics following saccadic eye movements.
We analyzed electrophysiology data recorded from silicon electrode arrays in V1 of free-viewing marmosets. Marmosets were shown a battery of artificial and natural stimuli with no fixation constraints: including sparse noise, spatiotemporal gabor noise, flashed gratings, and static natural images. In all cases, the marmosets had no fixation constraints, although oOne fixation condition was included where whitened natural images were rapidly updated at 30Hz. We fit 4-layer CNNs to combinations of stimulus conditions. The architecture in question has spatiotemporal 3D convolutions in the first layer. All subsequent layers are spatial only. Each layer was separated by a ReLU nonlinearity and batch normalization. The mapping from the convolutional layers to firing rate is captured by a readout layer that factorizes spatial position in the convolutional output and feature tuning. All parameters were fit end-to-end by minimizing the Poisson loss using the Adam Optimizer. We found that this architecture, trained on free-viewing natural images and gabor noise, captured the saccade-driven transients in V1 on witheld natural images and generalized to flashed images during fixation. In contrast, a CNN trained only on gabor noise did not generate saccade-driven transients and generalized less well to the fixation condition. Additionally, simple LN models did not produce transient responses, and did not match the performance of a vision-only CNN, even with the addition of saccade kernels. These results demonstrate that spatiotemporal CNNs can capture many of the response dynamics of V1 at high time resolution during natural oculomotor behavior.

Disclosures: D. Galor: None. **J. Mitchell:** None. **D.A. Butts:** None. **J.L. Yates:** None.

Poster

PSTR147. Visual Responses During Behavior I

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR147.05/Z7

Topic: D.06. Vision

Support: NIH Grant R01 EY030860
NIH Grant R01 NS120289

Title: Evidence for Bayesian-like computations in the mouse primary visual cortex during orientation discrimination

Authors: *P.-O. POLACK;
Rutgers Univ. - Newark, Newark, NJ

Abstract: We trust our sense to provide us with a reliable account of what is occurring in our surrounding environment. However, this impression of reliability is challenged by the existence of perceptual illusions such as the Necker cube or the Rubin's vase that have bi-stable percepts. In those situations, the switch between the two percepts can be prompted, suggesting that perception can be influenced by internal expectations. The idea of perception as an unconscious inference is centuries old. One of the most famous initial proponents of perception as an interaction between present sensations and previous experiences being Hermann von Helmholtz. More recently, this proposition was formalized in the Bayesian statistics framework: perception (posterior) can be modeled as the confrontation between expectation (prior) and sensory evidence (likelihood). Yet, it has been so far difficult to gather direct evidence that sensory processing follows this principle. Here, we report that in the primary cortex (V1) of mice performing a perceptual discrimination task, the representation visual stimuli can follow a dynamic transformation compatible with the Bayesian framework. During the presentation of the non-rewarded stimulus, the initial representation corresponds to the representation of the stimulus associated with the reward and therefore expected by the mouse (prior). This representation is progressively transformed into the representation of the non-rewarded stimulus while the mouse is exposed to the cue (likelihood). Finally, we found that the mouse uses the resulting representation to determine the behavioral outcome (posterior). Altogether, our results support the hypothesis that V1 performs Bayesian-like computations when processing visual evidence.

Disclosures: P. Polack: None.

Poster

PSTR147. Visual Responses During Behavior I

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR147.06/Z8

Topic: D.06. Vision

Support: NIMH ZIAMH002958

Title: Perceptual interpretation of optogenetic stimulation of inferior temporal cortex depends on the state of the visual system

Authors: *E. SHAHBAZI[1], T. MA[2], A. AFRAZ[3];
[1]NIH, Bethesda, MD; [2]New York Univ. Ctr. For Neural Sci., New York, NY; [3]NIH/NIMH, Bethesda, MD

Abstract: We have previously found that behavioral detection of cortical stimulation in the inferior temporal (IT) cortex depends highly on the images presented to the animals at the time of stimulation. The animals notice cortical stimulation easier while viewing some images and harder while looking at other images. We hypothesized that cortical stimulation in a given IT site does not lead to a constant perceptual effect independent of the visual input (e.g., a high-level phosphine); instead, cortical stimulation induces large perceptual changes for "easy images" compared to the hard ones. In order to put this hypothesis to the test, we appealed to perceptography. Perceptography is a novel method that utilizes high-throughput behavioral optogenetics and machine learning to graphically capture the visual perceptual events resulting from local stimulation in the macaque IT cortex. We trained two adult macaque monkeys to detect and report a brief optogenetic excitatory impulse administered to their central IT cortex through an implanted LED array. We then utilized machine learning to systematically warp a set of seed images to develop "Perceptograms". Perceptograms are specific images looking at which would trick the animal into reporting non-stimulated trials as stimulated (see previous work). To test our hypothesis, we first measured the animals' performance in detecting cortical stimulation while looking at seed images. As expected, there was a wide range of performance in the detection of cortical stimulation while fixating on various seed images. We then calculated the amount of image distortion in the perceptograms resulting from each seed image. The results showed a strong correlation ($r=0.71$, $p<0.0001$) between the ability to behaviorally detect brain stimulation while fixating on each seed image and the magnitude of the perceptual distortions in that image induced by optogenetic stimulation of the IT cortex. These results suggest that animals use stimulation-induced perceptual distortions to detect cortical stimulation. Moreover, these findings suggest that the perturbability of neural activity in the IT cortex highly depends on the state of other visual neurons, determined by the visual stimulus. This is a tantalizing possibility as it suggests that neural activity in IT lives on an intrinsic manifold, constraining the state of each neuron by the state of others.

Disclosures: E. Shahbazi: None. **T. Ma:** None. **A. Afraz:** None.

Poster

PSTR147. Visual Responses During Behavior I

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR147.07/Z9

Topic: D.06. Vision

Support: NIH NINDS R01NS109978

Title: Modifying visual perception by activating layer 4 (L4) excitatory neurons in primary visual cortex (V1)

Authors: *J. ZHUANG[1], J. DEL ROSARIO[1], S. COLETTA[2], J. AHN[1], B. HAIDER[3]; [1]Georgia Inst. of Technol., Atlanta, GA; [2]Emory Univ., Atlanta, GA; [3]Biomed. Engin., Georgia Tech. and Emory Univ., Atlanta, GA

Abstract: Direct electrical or optogenetic activation of primary visual cortex (V1) can lead to perceptual responses (Histed & Maunsell, 2014; Murphey & Maunsell, 2007), but several questions remain for understanding how "artificial percepts" arise. First, we do not know how direct V1 stimulation recruits excitatory and inhibitory neurons across layers, nor how this compares to laminar activity during normal visual percepts. Second, we do not know if direct stimulation could improve perceptual detection of otherwise weak visual stimuli.We investigated these questions in head-fixed, water-restricted mice expressing channelrhodopsin (ChR2) in V1 layer 4 (L4) while they performed a psychometric visual contrast detection task. We recorded neural activity with multisite linear silicon probes during task performance, and simultaneously optogenetically activated L4 neurons with focused laser light of varying intensities and durations. We assessed how putative excitatory and inhibitory neural activity evolved across cortical layers as a function of perceptual outcome, stimulus contrast, and laser intensity. Visual stimuli (static gratings) appeared in discrete regions of visual space, and we used intrinsic imaging to target recordings and laser stimulation (<0.3mm) to the retinotopically matched regions of V1. We calibrated laser parameters such that laser evoked spiking closely matched stimuli evoked spiking which were used during task performance. We found that V1 L4 stimulation significantly improved detectability of low contrast stimuli (18 ± 6% improvement for 1-5% contrast gratings; $p < 0.05$; 37 sessions in 2 mice). Perceptual responses also increased during L4 stimulation alone (>32 ± 11%). On trials of improved detectability, L4 regular spiking (RS) neuron firing rates also increased significantly within 0.2s following grating onset. Surprisingly, in some experiments laser stimulation reduced detectability of low contrasts, associated with relatively greater fast spiking (FS) putative inhibitory neuron activation. Exposing a control mouse (no opsin) to the same laser parameters did not alter performance (31 sessions in 1 mouse; $p > 0.05$ for all contrasts). Taken together, these preliminary findings suggest the effects of direct V1 stimulation on perception depend upon cell-type specific activation and recurrent cortical activity that can directly lead to changes in the sensitivity of visual perception. We are currently exploring how the relative activation of RS and FS neurons across layers predicts either enhanced or diminished detectability of visual stimuli.

Disclosures: **J. Zhuang:** None. **J. Del Rosario:** None. **S. Coletta:** None. **J. Ahn:** None. **B. Haider:** None.

Poster

PSTR147. Visual Responses During Behavior I

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR147.08/Z10

Topic: D.06. Vision

Support: NIH UF1 NS116377
AFOSR 19RT0316
Kavli Institute for Brain and Mind Postdoctoral Award

Title: The role of active vision in the primary visual cortex of freely-moving marmosets

Authors: *J. LI[1], V. SINGH[1], J. MITCHELL[2], A. HUK[3], C. MILLER[1];
[1]Dept. of Psychology, UCSD, SAN DIEGO, CA; [2]Brain and Cognitive Sci., Univ. of Rochester, Rochester, NY; [3]Psychiatry and Biobehavioral Sci., UCLA, Los Angeles, CA

Abstract: Historically, studies of visual cortex have been performed while nonhuman primates are head-fixed and viewing visual stimuli on a screen. This approach, however, is divorced from one of the primary factors that has driven the evolution of brains, including sensory systems, namely the ability to move through the world. In the real world, visual processing must accommodate how we actively explore the environment while making choices about when and where to go. Despite its clear significance, virtually nothing is known about how the primate visual system supports natural, active vision in freely moving animals. To address this problem, we leveraged an innovative, head-mounted eye-tracking system developed for marmosets in our lab while simultaneously recording the activity of single neurons in primate V1 to examine how the multiple sources of motion - eye, head, body - modulated neural activity and their effects on visual representations. In these experiments, monkeys are first head-fixed to quantify traditional visual stimuli for receptive field and tuning curve calculation, and natural scenes for comparison of the response properties from the same neurons while animals were freely-moving. During the freely-moving conditions, subjects are placed in a large arena where high-contrast visual stimuli are shown on the floor and wall. Analyses of data collected in the head-fixed condition show that we successfully recapitulated receptive fields and tuning curves of V1 neurons with our head-mounted eye-tracking system. Preliminary analyses of neural activity while marmosets were freely moving revealed a consistent and dominant saccade modulation of V1 activity, as well as modulation of neurons during head turns. While locomotion did modulate single neuron activity, its effects were the least pronounced of the sources of motion quantified here. The results suggest key differences in how self-motion signals impact V1 neural coding in primates as compared to rodents. Further work is being performed to elucidate the computational role of these different motor signals on V1 to support stable representations of the visual scene. These data are the first to examine the neural basis of active vision in a freely-moving primate and have a significant influence on our conceptions of natural vision.

Disclosures: J. Li: None. **V. Singh:** None. **J. Mitchell:** None. **A. Huk:** None. **C. Miller:** None.

Poster

PSTR147. Visual Responses During Behavior I

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR147.09/Z11

Topic: D.06. Vision

Support: NIH UF1 NS116377
NIH R01 NS118457

Title: Behavioral correlates of active vision in freely moving marmosets using head mounted eye-tracking

Authors: *V. SINGH, J. LI, C. MILLER;
UCSD, San Diego, CA

Abstract: Most of the vision neuroscience in primates has been carried out in highly controlled settings where the animal is head-fixed and restrained in a small volume. While this has the experimental advantage of allowing controlled presentation of stimulus and collection of large datasets with clear onset of cue and reliable behavioral data collection, it also significantly limits the visual experience of the animal, and by extension what we can learn about the neural basis of natural primate vision meaning that at present we know remarkably little about how primates naturally 'see' the world. Here we sought to bridge this gap by developing an innovative high-speed high resolution eye tracking system called CEREBRO (Chair-free Eye Recording using Backpack mounted Microcontrollers) for freely-moving marmoset monkeys. Using CEREBRO we have been able to perform reliable eye tracking in freely behaving common marmosets while also tracking the animals' head and body movements (using motion tracking cameras: Optitrack; and IMUs). This system also integrates seamlessly with wireless neural recording methods for freely-moving marmosets allowing for pioneering studies into the neural basis of natural active vision in primates. Analysis of marmoset visual behavior using CEREBRO revealed several novel findings that have significant implications for our conceptions of vision in primates. First, marmoset eye-movements when animals are head-fixed is robustly different from when animals are freely-moving, including both contexts in which the animal is locomoting and sitting while visually scanning the scene. Specifically, saccadic eye movements in freely-moving monkeys were shorter and more frequent than when head-fixed. Second, we discovered that almost all of the eye movements were in conjugation with head movements when marmosets are freely-moving. Given the smaller size of their head, marmosets moved their head in a saccade-fixation like pattern with eyes helping stabilize and refine these ballistic head shifts using compensatory movements providing novel insight into the motor mechanisms supporting stable visual scene representations. Overall, this innovative technology will provide a powerful new platform for studying the perceptual and neural basis of active vision during ethologically relevant behaviors in non-human primates.

Disclosures: V. Singh: None. **J. Li:** None. **C. Miller:** None.

Poster

PSTR147. Visual Responses During Behavior I

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR147.10/Z12

Topic: D.06. Vision

Title: Connectome-constrained continuous ring attractor networks of the fly compass system

Authors: *T. BISWAS[1], A. STANOEV[2], S. ROMANI[2], J. FITZGERALD[3];
[1]Janelia research campus, HHMI, Ashburn, VA; [2]Janelia Res. Campus, Ashburn, VA; [3]HHMI Janelia Res. Campus, Reston, VA

Abstract: A cognitive compass is a critical component of spatial navigation and path integration. Many previous models have used ring attractors to describe head direction systems. More recently, an accurate way to encode angular locations continuously with a relatively few neurons, as relevant for various insects, has also been designed. However, typically ring attractor models have relied on hand-designed neural network connectivity patterns, and we lack a principled methodology for quantitatively relating them to connectomics data. Here we introduce a novel theoretical framework to analyze the space of threshold-linear ring attractor networks that admit a continuous encoding of an angular coordinate. We show that to encode angles continuously with a (small) finite number of neurons the network must admit steady state *asymmetric* bumps of neural activity. Under the assumption that neural activity in the compass system is self-sustained, we derive predictions that link asymmetric bumps to symmetric patterns of neural network connectivity. We then provide a recipe for using neural activity measurements and connectomics data to test these predictions in the fly's compass system. Specifically, we examine the compass (EPG) neurons that form a ring-like structure in the fly central complex and exhibit bump-like activity encoding the fly's angular coordinate. The bump is believed to be sustained by a combination of direct local excitation between the EPGs and distal inhibition possibly mediated indirectly by Δ7 neurons. We asked whether one can find proportionality factors that convert measured synapse counts to synapse strengths such that the resulting network is a continuous ring attractor. Using symmetrized version of the measured synapse count matrices and four proportionality factors associated with EPG ↔ EPG, EPG ⟶ Δ7, Δ7⟶ EPG and Δ7 ↔ Δ7 synapses, we were indeed able to construct continuous attractor networks. Our theory was then able to predict the stable bump profiles that were found to be consistent with our fly activity measurements in the EPG's. Interestingly, we found that if we choose synapse count matrices randomly, approximately 15% of the time one could find proportionality factors that lead to a ring attractor. On one hand, this is encouraging as the measured connectome had to be in the right corner of the parameter space for our mechanism to be viable. On the other, it suggests that by just changing the proportionality factors one may be able to deal with small changes to the connectome. To summarize, we have provided a framework to incorporate connectomics data and build data-driven ring attractor models that are close to biology.

Disclosures: T. Biswas: None. **A. Stanoev:** None. **S. Romani:** None. **J. Fitzgerald:** None.

Poster

PSTR147. Visual Responses During Behavior I

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR147.11/Z13

Topic: D.06. Vision

Support: ECAE Research Grant (RP-255-2023)
ASPIRE-AARE grant

Title: Testing the inhibition hypothesis for EEG alpha rhythm in visual detection: preliminary findings

Authors: *K. SUNG[1], M. ALJAAFARI[2], N. AL AMERI[2], C. HABAK[2];
[1]Emirates Col. For Advanced Educ., Abu Dhabi, United Arab Emirates; [2]Emirates Col. for Advanced Educ., Abu Dhabi, United Arab Emirates

Abstract: The alpha (8 - 13Hz) EEG oscillation has received much attention, as it is firmly correlated with cortical activities at different levels of human information processing (e.g., sensation, attention, & memory). In healthy individuals, alpha power is attenuated (desynchronization) in cortical areas related to goal-directed and task-related neuronal activities, and enhanced (synchronization) in sites unrelated to a task. Some have suggested that alpha oscillation represents the electrophysiologic mechanism that controls neuronal activities: a mechanism that activates/suppresses cortical activities, pointing to causality. We aim to test the causality claim (i.e., inhibition hypothesis), hypothesizing that if it held, alpha power would be higher in the high noise condition (difficult) than in the low or no noise (easy) conditions, because of suppression of the noise-related neuronal activity. Six adults (2 males) participated in an EEG experiment: they viewed a movie consisting of a baseline blank screen (2 s), followed by static random noise (1.6, 1.8, 2.2 s), which then remained on-screen while a static Gabor target (4 angles) gradually increased in intensity (3.2 s), to a maximum (1.6 s), then gradually decreasing (3.2 s), and the noise remained (2 s). Participants were asked to press a key to indicate the appearance and disappearance of the target. Noise level was manipulated through luminance modulation and target luminance was constant across conditions. Trials with different noise levels were randomly intermixed into three blocks of trials (36 trials/block). A specialized monitor (VIEWPixx/EEG) was used to generate movie frame-based event markers, and EEG signals were recorded using a 64-channel high-impedance EEG system (R-net with actiChamp Plus, Brain Products). Results from three sites (Oz, Pz, & Fz) showed that on average, alpha power attenuated at the onset of the noise, with no power difference across conditions. The difference in alpha power attenuation was observable between conditions only when the target was seen, with the largest power reduction in the difficult condition. The event-related spectral perturbation showed lasting alpha power reduction across all time windows in the difficult condition, but not in easy conditions: alpha power recovered to almost baseline levels when the target was at maximum intensity in easy conditions, but it remained reduced in the difficult condition. These tentative results are not in line with the causality claim, but analysis across other sites and additional participants are needed for conclusive evidence.

Disclosures: K. Sung: None. **M. Aljaafari:** None. **N. Al Ameri:** None. **C. Habak:** None.

Poster

PSTR147. Visual Responses During Behavior I

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR147.12/Z14

Topic: D.06. Vision

Support: NIH Grant EY030864

Title: Role of light exposure and melanopsin in exploration versus aversion

Authors: *R. GOIT, S. PARIKH, A. MATYNIA;
UCLA Stein Eye Inst., Los Angeles, CA

Abstract: Behavioral states can be altered by light, and the melanopsin trigeminal and retinal circuitry have been shown to mediate responses to light. Whether light is experienced as positive (mood) or negative (pain or discomfort) can also be influenced by pathophysiology. For example, photoallodynia is characterized by light aversion and the perception of minor irritation to debilitating pain. The aim of this study was to identify mechanisms of light-avoiding versus light-seeking behavior, including any role for the melanopsin system. Melanopsin knockout ($OPN4^{-/-}$) mice and wild type litter mate controls ($OPN4^{+/+}$) of both sexes were used. Mice were exposed to no light or 30 min/day of light (1000 lx) for 30 days. Exploratory drive and light avoidance were assessed using a light/dark box with an overhead LED lighting system. Corneal mechanical sensitivity was assessed using the Von Frey Filaments test. Corneal innervation was measured using immunofluorescence for b-tubulin in corneal whole mounts. Quantitative polymerase chain reaction was used to measure the gene expression in the trigeminal ganglion. Bright light exposure and habituation increases the drive to explore the open chamber at both 0 and 1000 lux. This effect is independent of melanopsin, but prior light experience influences exploratory drive versus light avoidance in a melanopsin-dependent manner. Corneal mechanical sensitivity and innervation were comparable between the genotypes and treatment. Preliminary data indicates that gene expression of transient receptor potential vanilloid type 1 (TRPV1) was significantly lower in $OPN4^{-/-}$ mice after light exposure and was similarly lower but did not reach significance in unexposed $OPN4^{-/-}$ mice. Together these data suggest that prior light exposure influences exploratory drive, acts at least partially via melanopsin, and does not increase pain sensitivity.

Disclosures: R. Goit: None. **S. Parikh:** None. **A. Matynia:** None.

Poster

PSTR148. Higher Visual Areas

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR148.01/Z15

Topic: D.06. Vision

Support: NIH R01EY033835
NSF GRFP DGE-1656518

Title: White matter connectivity of high-level visual areas is organized by cytoarchitecture from birth

Authors: *E. KUBOTA[1], X. YAN[2], S. TUNG[1], B. FASCENDINI[3], M. GROTHEER[4], K. GRILL-SPECTOR[1];
[1]Stanford Univ., Stanford, CA; [2]Inst. of Sci. and Technol. for Brain-Inspired Intelligence, Shanghai, China; [3]Princeton Univ., Priceton, NJ; [4]Dept. of Psychology, Philipps-University Marburg, Marburg, Germany

Abstract: The spatial organization of high-level visual areas is consistent across individuals, which has led researchers to ask about the factors that constrain their organization. One hypothesis is that white matter connectivity provides a scaffold upon which functional selectivity emerges. In particular, it has been hypothesized that white matter connectivity might be an innate or early developing constraint on the development of later function (Li et al., 2020; Mahon & Caramazza, 2011). In past work, we found that white matter connections of high-level visual areas identified in individual participants were organized by cytoarchitecture rather than category-selectivity in childhood, and by both cytoarchitecture and category-selectivity in adulthood (Kubota et al., 2023). In the present study, we tested whether the relationship between cytoarchitecture and white matter was present from birth. Using diffusion magnetic resonance imaging (dMRI) we measured white matter connectivity of high-level visual areas in 68 infants (N(newborns)=23, 8 females, meanSD age: 28.610.2 days), N(3-months)=25, 10 females, mean SD age: 105.617.3 days, and N(6-months)=20, 10 females, mean SD age: 187.213.5 days). To do so, we projected six maximum probability maps of functional regions of interest (fROIs) from adult participants to infant brains using cortex based alignment. Using these fROIs, we find that white matter connectivity is more similar for regions within the same cytoarchitectonic area (β = 0.58, $p < 2x10\text{-}16$), with a smaller effect of category ($\beta = 0.08$, $p = 0.04$) for all three age groups (no significant effect of age). Next, we smoothly tiled the cortical surface with equally spaced disks and measured the organization of white matter connectivity independently of category-selectivity to test whether there are sharp boundaries or a smooth gradient in white matter connectivity. We find that disks located in the same cytoarchitectonic area have more similar connectivity, even when adding distance as a covariate, reflecting sharp boundaries in connectivity. Together, these results suggest that regions located within the same cytoarchitectonic area have similar white matter connectivity from birth.

Disclosures: E. Kubota: None. **X. Yan:** None. **S. Tung:** None. **B. Fascendini:** None. **M. Grotheer:** None. **K. Grill-Spector:** None.

Poster

PSTR148. Higher Visual Areas

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR148.02/Z16

Topic: D.06. Vision

Support: H2020-MSCA-IF-2020/MSCA-IF-GF Grant 101025482

Title: Deep learning synthesis of optimal stimuli across mouse lateral visual hierarchy

Authors: *M. DIAMANTAKI[1], K. F. WILLEKE[2], S. S. PATEL[3], K. FRANKE[3], F. SINZ[4], A. S. TOLIAS[3], E. FROUDARAKIS[1];
[1]Inst. of Mol. Biol. and Biotech., Fndn. for Res. and Technology, Hellas, Heraklion, Greece; [2]Werner Reichardt Ctr. For Integrative Neurosci., Tuebingen, Germany; [3]Dept. of Neurosci., Baylor Col. of Med., Houston, TX; [4]Univ. of Göttingen, Göttingen, Germany

Abstract: Invariant object recognition is the ability of animals to rapidly recognize objects irrespective of variations in their appearance. This remarkable ability is mediated by the ventral visual stream in primates, a set of hierarchically organized interconnected visual areas. To identify the computational role of neurons along the ventral stream, deep neural networks have been routinely used in primates, due to their hierarchical organization that resembles the organization of the ventral stream. In mice, anatomical and physiological studies have revealed a network of lateral higher-order cortical visual areas (HVAs) which are believed to form the mouse ventral visual stream. While deep learning approaches have explored optimal stimuli in mouse visual areas, similar to primate studies, they have mostly been restricted to functional imaging data from the primary visual cortex (V1). Here, we seek to expand these approaches in mice by using large-scale electrophysiological data from multiple visual cortical areas to generate a digital twin of the mouse visual cortex. The digital twin model allowed us to synthesize optimal visual stimuli that maximally drive the neurons (most exciting inputs - MEIs). Specifically, by using the Neuropixels probes we simultaneously recorded the activity of hundreds of neurons in vivo in mouse V1 and lateral HVAs in response to natural images. We then trained a convolutional neural network to predict the responses of each neuron recorded across the different areas and generated a set of MEIs that would optimally excite the recorded neurons. Subsequently, we showed the optimized stimuli back to the mice and recorded the activity of the same neurons. This enabled us to verify in vivo the results of the in silico model predictions. Additional investigations of these closed-loop electrophysiological data from thousands of neurons are ongoing to uncover neural invariances across the visual hierarchy, and to disentangle invariant directions in the stimulus space. Identifying the neural invariances across lateral HVAs will then allow us to dissect the role of hierarchical processing in complex cortical computations such as invariant object recognition.

Disclosures: **M. Diamantaki:** None. **K.F. Willeke:** None. **S.S. Patel:** None. **K. Franke:** None. **F. Sinz:** None. **A.S. Tolias:** None. **E. Froudarakis:** None.

Poster

PSTR148. Higher Visual Areas

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR148.03/Z17

Topic: D.06. Vision

Support: NSERC and CIHR to CC.

Title: Enhanced Sensitivity to Precision in Cortical Visual Area 21a

Authors: *L. IKAN[1], N. CORTES[1], H. LADRET[1,2], L. PERRINET[2], C. CASANOVA[1];
[1]Univ. de Montreal, Montreal, QC, Canada; [2]Inst. des Neurosciences de la Timone, Aix-Marseille, France

Abstract: Our visual environment is composed of distributions of variable orientation precisions. Here we focused on understanding how the precision of visual stimuli affects the processing of orientation in higher hierarchical areas. Specifically, we examined cortical area 21a in cats, often considered the equivalent of primate area V4 within the hierarchical organization of visual processing. Using pseudo-natural visual stimuli called "MotionClouds," (MC) we investigated the impact of orientation precision on the responses of neurons in area 21a. Four parameters control the MC precision: the orientation, the spatial frequency (SF), B_{θ} and B_{sf}. The latter two regulate the bandwidth of the orientation and SF, respectively. We recorded and analyzed responses of 21a neurons to determine variations in orientation precision. Preliminary data reveal that neurons in area 21a exhibit mainly two types of responses to MC. The first type is a binary response, where neurons show strong activation to high-precision stimuli (B_{θ} low) but do not respond to low-precision stimuli (B_{θ} high). The second type of response demonstrates a gradual decrease in neural activity as precision decreases. Interestingly, both responses maximize their activity to different SFs to those used to maximize the response to sinusoidal drifting gratings. This data suggest the involvement of the cortical ventral stream in precision processing. However, a key question remains: how does the cortex utilize this precision information to shape visual perception?
Supp: NSERC and CIHR to CC.

Disclosures: L. Ikan: None. **N. Cortes:** None. **H. Ladret:** None. **L. Perrinet:** None. **C. Casanova:** None.

Poster

PSTR148. Higher Visual Areas

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

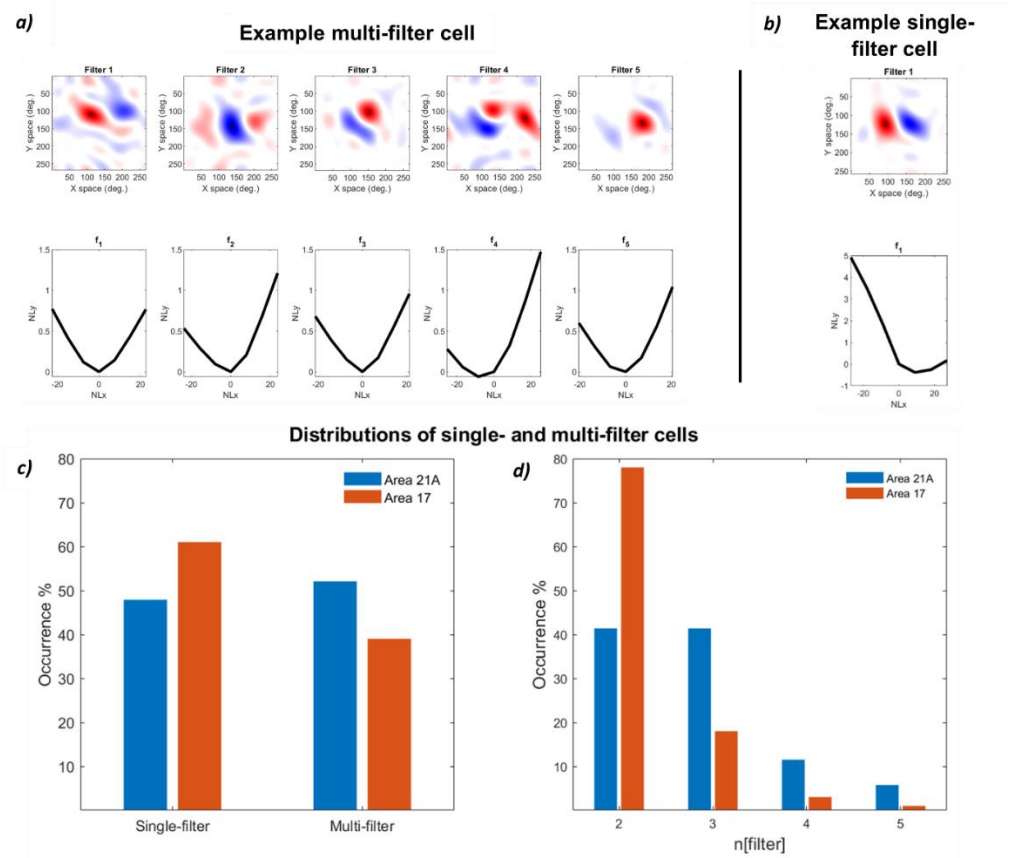
Program #/Poster #: PSTR148.04/Z18

Topic: D.06. Vision

Title: Properties of neural receptive fields in area 21A of the cat cortex.

Authors: *J. C. SIBBERAS[1], H. MEFFIN[1], M. R. IBBOTSON[2], J. JUNG[2];
[1]The Univ. of Melbourne, Melbourne, Australia; [2]Natl. Vision Res. Inst., Melbourne, Australia

Abstract: Object recognition in the cortex is enabled by extraction of increasingly complex features from the visual field. The function of various late-stage processing areas in the ventral pathway has been described phenomenologically, such as face recognition in regions of the inferotemporal cortex, and the position-invariant selectivity for shape in V4. However, a mechanistic understanding of visual information processing, such as through receptive field (RF) analysis and parametric modelling is in its infancy beyond V1. To extend our understanding of processing in the ventral pathway beyond these areas, we recorded activity from cells in area 21a (homologous to primate V4) during presentation of natural image stimuli and investigated neural RF structures and feature-response relationships. We fit receptive fields to 167 cells using the nonlinear input model (NIM). Neurons in area 21A exhibited a range of differences to that of area 17. For instance, RFs in this area are composed of a mix of oriented Gabor-like filters and bloblike receptive fields that are larger and more complex than those seen in area 17 (homologous to V1) and are less distinctly classified. The average length and width for area 21A receptive fields was 9° and 6° respectively, where area 17 cells are typically in the range of 1-6°. Interestingly, a subset of area 21A cell RFs extended across the full range of the stimulated visual field, over 21°. Further, NIM RFs for neurons in area 21A are more commonly multi-filter (51%, n=167) than that of area 17 (39%, n=192), suggesting not only a more complex range of feature selectivity but a more complex ability to perform nonlinear processing of these features. In turn, we are beginning to illustrate how area 21A neurons extract more complex features of the visual field than those of primary visual cortex neurons and play a role in hierarchical processing of form and structure in vision.



Disclosures: **J.C. Sibberas:** None. **H. Meffin:** None. **M.R. Ibbotson:** None. **J. Jung:** None.

Poster

PSTR148. Higher Visual Areas

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR148.05/Z19

Topic: D.06. Vision

Support: DFG Grant
CIRC Grant

Title: Segregated divisions of macaque lateral intraparietal area (LIP) revealed by post-mortem diffusion-weighted imaging and tractography

Authors: *M. WIRSUM[1,2], J. E. SMITH[3,4], T. B. DYRBY[5,6], K. KRUG[1,2,3];
[1]Dept. in Sensory Physiol., Institue of Biology, Otto von Guericke Univ., Magdeburg, Germany; [2]CIRC, Deutsches Zentrum für psychische Gesundheit, Magdeburg, Germany; [3]Dept. of Phys., Anat. & Gen., Univ. of Oxford, Oxford, United Kingdom; [4]Ernst Strüngmann Inst. for Neurosci. with Max Planck Society, Frankfurt, Germany; [5]Danish Res. Ctr. for MR, Hvidovre, Denmark; [6]Dept. of Applied Mathematics and Computer Science, Tech. Univ. of Denmark, Lyngby, Denmark

Abstract: Lateral intraparietal area (LIP) plays a central role in the sensorimotor-transformations for targeting eye movements and spatial attention. Anatomically, LIP consists of two distinct regions: a heavily myelinated ventral region (LIPv) and a sparsely myelinated dorsal region (LIPd). LIPv and LIPd are associated with different functions and differ in their connectivity patterns with other brain areas. For now, these subdivisions can only be clearly delineated with histological approaches.
We investigated whether LIP can be divided into LIPd and LIPv based on their differential connectivity profiles with other areas, using high resolution diffusion weighted imaging (DWI) data and probabilistic tractography obtained from 4 *post mortem rhesus macaque* brains (2 females, 2 males, mean age = 8.75 ± 3.20 years). After systematic analysis of histological data from 25 publications, evaluating 39 potential target areas, six areas were chosen for tractography because of their robust connectivity with predominantly one of the two LIP regions: dorsal visual stream areas V3A, V6A and V5/MT, primarily connected to LIPv, and ventral visual stream areas IPa, TEm and V4d, mainly connected to LIPd. Tractography was run using *FSL* on the pre-processed, diffusion-modelled data to estimate the white matter tracts from LIP to the targets. Based on the relative strength of connectivity to these targets, LIP was then hardsegmented. Qualitatively, LIP was divided in 6 out of 8 hemispheres into a more dorsal and more ventral part, which overlapped to a large extent with the ground-truth histological D99 atlas. When individual maps were randomly permuted 10,000 times, 7 out of 8 predicted maps performed better than chance ($p < 0.025$). The mean match with ground-truth for LIPv was 74.7% (STD = 8.9 %) and for LIPd 62.0% (STD = 21.2 %).
Excluding targets in individual hemispheres based on anatomical plausibility and uncertainty of estimated white matter tracts improved the overlap with the ground-truth LIPd/LIPv division in

the D99 atlas with a mean match of 79.6% (STD = 20.0 %) for LIPv and 67.1% (STD = 18.8 %) for LIPd (n=8). All 8 maps performed better than chance ($p < 0.025$).
We show that non-invasive DWI with probabilistic tractography can reliably segment parietal area LIP and potentially other cortical areas into their structural subdivisions in individual subjects based solely on connectivity. As next step, this approach is being applied to *in vivo* macaque and human brains to define subregions and circuits in individual subjects for research and clinical applications.

Disclosures: **M. Wirsum:** None. **J.E. Smith:** None. **T.B. Dyrby:** None. **K. Krug:** None.

Poster

PSTR148. Higher Visual Areas

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR148.06/Z20

Topic: D.06. Vision

Support: R01MH111417

Title: Selectivity and temporal dynamics of neural responses in category-selective cortex are associated with differences in spatial tuning

Authors: L. DIETZ[1], A. BRANDS[1], G. PIANTONI[3], S. MONTENEGRO[4], A. FLINKER[4], S. DEVORE[4], O. DEVINSKY[4], W. DOYLE[4], P. DUGAN[4], D. FRIEDMAN[4], N. RAMSEY[3], N. PETRIDOU[3], J. WINAWER[5], **I. I. A. GROEN**[2,5];
[1]Informatics Inst., [2]Informatics Institutde, Univ. of Amsterdam, Amsterdam, Netherlands; [3]Univ. Med. Ctr. Utrecht, Utrecht, Netherlands; [4]New York Univ. Grossman Sch. of Med., New York, NY; [5]Dept. of Psychology, New York Univ., New York, NY

Abstract: Extrastriate visual cortex contains neural populations that exhibit selectivity to specific stimulus classes (e.g., scenes vs. faces) that are spatially clustered in specific anatomical locations (e.g., parahippocampal vs. fusiform gyrus). One account of this stereotyped organisation is that it reflects different visual processing demands, with face processing requiring detailed analysis of foveated visual input, and scene processing requiring large scale analysis of the global visual field. Indeed, fMRI studies suggest a coupling between category preference and spatial tuning, with face-selective regions having smaller, foveally-centred population receptive fields (pRFs) and scene-selective regions having larger, peripherally-extending pRFs. Here, we confirm this association in electrophysiological recordings in human visual cortex. We analysed ECoG data (https://openneuro.org/datasets/ds004194) containing broadband responses to traversing bar sequences, allowing for the estimation of population receptive fields, as well as responses to centrally presented face, house and scene stimuli, allowing measurement of category-selectivity. By aggregating data across multiple participants (n=6, both sexes) we identified 55 electrodes that exhibited significant face or scene/house selectivity ($d' > 0.5$) whilst also showing robust pRF fits (cross-validated $R2 > 20\%$). Comparison of pRF properties and

selectivity profiles shows that face- and scene/house-selective electrodes differ in pRF characteristics, with scene/house-selective electrodes having more peripheral and larger pRFs than face-selective electrodes. A regression model using pRF eccentricity and size as regressors explains 28% of the variance in face vs. scene/house selectivity. Capitalising on the high temporal resolution of ECoG, we also explored the association between pRF properties and temporal dynamics of neural responses to category stimuli. Increased pRF sizes are associated with slower rise and slower decay of responses to face and scene/house stimuli. Across electrodes, pRF properties explained 26-38% of the variance in the time to peak and rate of response decay of face and scene/house responses. Although pRF properties exhibited associations with both selectivity and temporal dynamics of face and scene/house responses, we find no clear direct relation between stimulus selectivity and temporal dynamics. These results extend fMRI observations of a link between spatial tuning and category preference of neural population responses in human visual cortex, and additionally demonstrate a separate influence of spatial tuning on neural response dynamics.

Disclosures: **L. Dietz:** None. **A. Brands:** None. **G. Piantoni:** None. **S. Montenegro:** None. **A. Flinker:** None. **S. Devore:** None. **O. Devinsky:** None. **W. Doyle:** None. **P. Dugan:** None. **D. Friedman:** None. **N. Ramsey:** None. **N. Petridou:** None. **J. Winawer:** None. **I.I.A. Groen:** None.

Poster

PSTR148. Higher Visual Areas

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR148.07/Z21

Topic: D.06. Vision

Title: Probing the role of cortical specialization in visual spatiotemporal processing

Authors: *S. CETINDAG[1,2,3], Y. ACIR[1], B. VERMAERCKE[1,3], A. AYAZ[1,3], V. BONIN[1,2,4,3]; [1]Neuro Electronics Res. Flanders (NERF), Leuven, Belgium; [2]Dept. of Biol., KU Leuven, Leuven, Belgium; [3]VIB, Leuven, Belgium; [4]imec, Leuven, Belgium

Abstract: The mammalian visual cortex is comprised of a network of functionally specialized higher visual areas (HVAs) that encode distinct aspects of the visual scene. Although it is hypothesized that this specialization could predict the contributions of certain areas, the impact of cortical specialization in recruiting HVAs to visual perception is unclear. We devised a reaction time task to characterize the link between activity in specialized HVAs and the perception of stimuli of different spatial (SF) and temporal frequencies (TF) in the mouse. GCaMP6 expressing Ai32ChR2 x PVCre mice were trained to detect a narrowband spatiotemporal noise stimulus (0.04cpd-8Hz) presented on the left- or right-hand side of the visual field. Areas V1 and individual HVAs were identified using widefield calcium imaging. To probe the role of visual areas, V1 and HVA activity was inhibited by selective activation of PV neurons with patterned-light optogenetics using a digital micromirror device (DMD). Consistent

with a role of visual cortical activity, inhibiting activity in V1 or HVAs impaired the detection of both low and high contrast stimuli (0.04cpd-8Hz, 12.5% contrast V1: hit rate 81 ± 7% vs 16 ± 4%, $p<0.01$ Mann-Whitney U test N=5 mice; HVAs: hit rate 81 ± 3% vs 17 ± 2%, $p<0.05$, 50% contrast V1: hit rate 96 ± 3% vs 76 ± 11%, $p<0.05$, N=5; HVA: hit rate 98 ± 1% vs 57 ± 11%, $p<0.05$ N=4) and increased response times (0.04cpd-8Hz, 12.5% contrast V1: response time 519 ± 71ms vs 1012 ± 3ms, $p<0.05$ Mann-Whitney U test N=5; HVAs: response time 539 ± 49ms vs 1011 ± 4ms, $p<0.05$ N=4; 50% contrast V1: response time 289 ± 18ms vs 615 ± 104ms, $p<0.01$, N=5; HVAs: response time 307 ± 20ms vs 785 ± 131ms, $p<0.05$ N=4). Inhibition of individual HVAs yielded area-specific effects with more pronounced impairment when inhibiting lateral areas (LM, AL, RL) than medial areas (PM, AM) (12.5% contrast Δ hit rate = 30% vs 13%, $p<0.001$ Mann-Whitney U Test, N=5). Additionally, we used the assay to examine the link between the tuning of HVAs and their involvement in visually guided behavior. In preliminary experiments, consistent with its preferred tuning, suppression of AL activity during the presentation of low SF and high TF stimuli (0.04cpd-8Hz) yielded more pronounced effects than high SF and low TF stimuli (0.16cpd-0.5Hz) (12.5% contrast, 0.16cpd-0.5Hz, hit rate 53 ± 9% vs 40 ± 10%, p=0.6 Mann-Whitney U test; 0.04cpd_8Hz, hit rate 85 ± 3% vs 54 ± 2%, p=0.3 N=2). The results of this study will reveal the engagement of HVAs in processing diverse spatiotemporal visual information, providing new insights into the relationship between the functional properties of neuronal populations and their contribution to visual perceptual behavior.

Disclosures: S. Cetindag: None. **Y. Acir:** None. **B. Vermaercke:** None. **A. Ayaz:** None. **V. Bonin:** None.

Poster

PSTR148. Higher Visual Areas

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR148.08/Z22

Topic: D.06. Vision

Support: NIMH grant 1R01MH127199-01A1
NSF grant 1835200 (MIG)

Title: Representational geometries supporting social action understanding in natural vision

Authors: *J. HAN[1], S. A. NASTASE[2], M. GOBBINI[3], J. HAXBY[1];
[1]Dartmouth Col., Hanover, NH; [2]Princeton Univ., Princeton, NJ; [3]Univ. of Bologna, Bologna, Italy

Abstract: Humans have a remarkable ability to understand the actions and intentions of others by observing their behavior. To investigate how the cortical processing hierarchy supports action understanding, we conducted a study using functional magnetic resonance imaging (fMRI) to measure brain activity while participants watched a variety of real-life action clips, both social

and non-social. We analyzed the data using representational dissimilarity matrices (RDMs) capturing different hypothesized feature spaces related to visual and action understanding. The RDMs included measures of motion energy, gaze trajectories, semantic content (verbs and non-verbs), and behavioral judgments based on multiple arrangement tasks. These behavioral tasks involved arranging the video clips according to their social or goal-directed (transitivity) content, as well as arranging static images representative of each clip based on the similarity of depicted objects, people, or scenes. Using searchlight representational similarity analysis, we examined neural representational geometries across the cortical processing hierarchy. Our results revealed that semantic content and behavioral judgments accounted for more variance in brain activity than low-level visual features. Specifically, these factors showed stronger effects in the lateral occipitotemporal, ventral temporal, inferior parietal, and premotor cortex, which are brain regions involved in action observation and understanding. Comparing the multiple arrangement tasks, we found that behavioral judgments of sociality and transitivity in the video clips better captured the neural representational geometries than the static-image arrangement tasks. The representational geometries based on sociality and transitivity judgments outperformed other RDMs, including verb semantics, in predicting neural RDMs. This suggests that subjective behavioral judgments of similarity capture unique attributes of neural representation that are not captured by well-studied feature-based descriptions. Overall, the representational geometry of transitivity — action goals — better predicted neural representational geometry in ventral temporal, inferior lateral occipital, parietal and premotor cortices, whereas the representational geometry of sociality better predicted neural representational geometry in the precuneus, more superior lateral occipital cortex and the posterior superior temporal sulcus. Our findings suggest that the neural system for action perception primarily represents the meaning of human actions and that the representations of action goals and social content are dissociable.

Disclosures: **J. Han:** None. **S.A. Nastase:** None. **M. Gobbini:** None. **J. Haxby:** None.

Poster

PSTR148. Higher Visual Areas

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR148.09

Topic: D.06. Vision

Support: SPP 2205 Evolutionary optimization of neuronal processing

Title: A three-dimensional functional investigation of the avian visual cortex - Do birds share a canonical forebrain circuit?

Authors: *W. CLARK[1], C. S. SEVINCIK[2], D. RÖDERS[3], W. SEAH[3], O. GÜNTÜRKÜN[2], J. ROSE[3], R. PUSCH[2];
[1]Fac. of Psychology, Ruhr-Universität Bochum: Ruhr-Universitat Bochum, Bochum, Germany; [2]Biopsychology, [3]Neural Basis of Learning, Inst. for Cognitive Neurosci., Bochum, Germany

Abstract: Birds display outstanding visuo-cognitive capabilities, yet the avian visual forebrain displays a nuclear neuronal organization with no direct equivalent of the ventral visual stream. Despite these differences, birds may code for higher-order features using similar principles to the hierarchal arrangement of layers found in the mammalian visual cortex. To establish the pigeon as a model for higher-order visual processing, we analyzed the receptive field organization of neurons in a stack of forebrain territories known as the entopallium, nidopallium, and mesopallium with sparse noise stimuli using 256 channel silicon electrodes. We found evidence that response latency, selectivity, and receptive field organization of neurons within all three visual forebrain structures may differ across the 3-D volume of the visual system that we targeted. In addition, the phase of local field potential oscillations rapidly reversed at sites corresponding to the thalamic input zone of the entopallium. These findings are consistent with the notion that these structures contribute to a global analysis of the visual scene and might process information in a hierarchical manner that is comparable with the mammalian visual cortex.

Disclosures: W. Clark: None. **C.S. Sevincik:** None. **D. Röders:** None. **W. Seah:** None. **O. Güntürkün:** None. **J. Rose:** None. **R. Pusch:** None.

Poster

PSTR148. Higher Visual Areas

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR148.10/Z23

Topic: D.06. Vision

Support: NIH Grant EY029438
Whitehall Foundation (2016-08-18)

Title: Hierarchical transformation of 3D object representations across macaque V3A and CIP

Authors: *R. DOUDLAH[1], L. KRESSER[1], T.-Y. CHANG[2], B. KIM[1], A. SUNKARA[3], A. ROSENBERG[1];
[1]Neurosci., Univ. of Wisconsin-Madison, Madison, WI; [2]Sch. of Med., Natl. Def. Med. Ctr., Taipei, Taiwan; [3]WiSys Technol. Fndn., Madison, WI

Abstract: The 3D world is perceived as stable, yet the 2D retinal projections of the scene depend greatly on gaze position and necessarily confound 3D position and orientation information. Where and how the primate visual system transforms ambiguous 2D retinal signals into 3D representations that are robust to the observer's gaze remain unclear. Recently, we found a hierarchical transformation from lower-level visual feature selectivity to higher-level 3D object pose (position and orientation) selectivity across macaque area V3A and the caudal intraparietal (CIP) area. In that work, 3D surface orientation selectivity was measured at different distances while the fixation distance (vergence) was held constant. Here, we tested if gaze position information is utilized within this hierarchy to stabilize 3D visual representations. Specifically,

we performed a complementary experiment in which 3D orientation selectivity was measured at different fixation distances (in front of, at, and behind the surfaces) while the stimulus distance was held constant relative to the monkey. In parallel, the monkeys performed an eight-alternative 3D orientation discrimination task in which they reported the tilt of the viewed plane (i.e., which side was closest). Intriguingly, we found that the responses of more neurons in V3A, the lower-level area, were modulated by the fixation distance during passive viewing of a small target, than in CIP. To quantify if the 3D orientation selectivity depended on the fixation distance, we assessed the separability of the joint 3D orientation and vergence tuning curves. Consistent with previous results in which the stimulus distance was varied but the fixation distance was constant, separability was greater in CIP than V3A. This indicates that the shape of 3D orientation tuning curves was more tolerant to vergence in CIP than V3A. These results are consistent with a hierarchical model in which a combination of lower-level visual signals and vergence signals at the level of V3A are used to compute higher-level 3D object orientation representations in CIP which are more tolerant to the moment-to-moment volatility of eye position. Also consistent with previous results, we found that choice-related activity during the 3D orientation discrimination task began earlier in V3A than CIP, was most prevalent in CIP, and was preferentially carried by higher-level 3D feature selective neurons. These findings collectively implicate V3A and CIP in the hierarchical transformation of volatile 2D retinal signals into stable, behaviorally relevant 3D object representations.

Disclosures: **R. Doudlah:** None. **L. Kresser:** None. **T. Chang:** None. **B. Kim:** None. **A. Sunkara:** None. **A. Rosenberg:** None.

Poster

PSTR148. Higher Visual Areas

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR148.11/Z24

Topic: D.06. Vision

Support: NIH Grant EY029438
Whitehall Foundation (2016-08-18)

Title: Saccade-related activity and sensorimotor associations at the macaque parieto-occipital junction

Authors: *L. KRESSER[1], R. DOUDLAH[1], Z. ZHU[1], T.-Y. CHANG[2], B. KIM[1], A. ROSENBERG[1];
[1]Univ. of Wisconsin - Madison Neurosci. Training Program, Madison, WI; [2]Sch. of Med., Natl. Def. Med. Ctr., Taipei, Taiwan

Abstract: Forming behaviorally relevant 3D visual representations of objects and implementing sensorimotor transformations allow for successful interactions with the environment. In macaque monkeys, these processes are supported by area V3A and the caudal intraparietal area (CIP),

which bridge the parieto-occipital junction. We previously found that high-level 3D representations and saccade-related signals occur in parallel across the V3A-to-CIP hierarchy. In that work, monkeys were trained to discriminate the orientation of 3D planar surfaces and perform a visually guided (pop-up) saccade task. That work revealed neurons in both areas whose activity predicted the direction and timing of saccades as well as a sensorimotor association in which the 3D orientation and saccade direction preferences generally aligned.To further investigate the saccade-related properties of V3A and CIP and assess if sensorimotor associations are inherent to the areas, we trained a monkey to perform pop-up, overlap, and memory-guided saccade tasks, as well as to view 3D oriented planar surfaces during passive fixation. In both the pop-up and overlap saccade tasks, the percentage of neurons with saccade direction selectivity was greater in CIP (pop-up: 84%; overlap: 72%) than V3A (pop-up: 59%; overlap: 45%). The overlap saccade task further attributed that selectivity to sustained activity preceding the eye movements, whose level depended on the saccade direction. This was confirmed by the memory-guided saccade task, in which direction selective sustained activity was found during the delay period in which no saccade target was present. The memory-guided saccade task further established the cross-area difference in the percentage of neurons that carried saccade-related activity, with 59% of CIP but only 35% of V3A neurons maintaining direction selectivity. Notably, although both areas carried saccade-related activity, neither showed clear evidence of ramping activity commonly associated with presaccadic signals. The results instead suggest that memory traces of target locations that inform oculomotor responses are hierarchically constructed across V3A and CIP.Consistent with previous work, we also found that the majority of neurons were selective for 3D surface orientation. As such, many carried both 3D orientation and saccade direction information. However, the orientation and saccade direction preferences of the neurons did not generally align. This suggests that the sensorimotor association previously observed in both areas following extensive training in a 3D orientation discrimination task were experience dependent.

Disclosures: **L. Kresser:** None. **R. Doudlah:** None. **Z. Zhu:** None. **T. Chang:** None. **B. Kim:** None. **A. Rosenberg:** None.

Poster

PSTR149. Functional Organization of the Developing and Mature Visual System

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR149.01/Z25

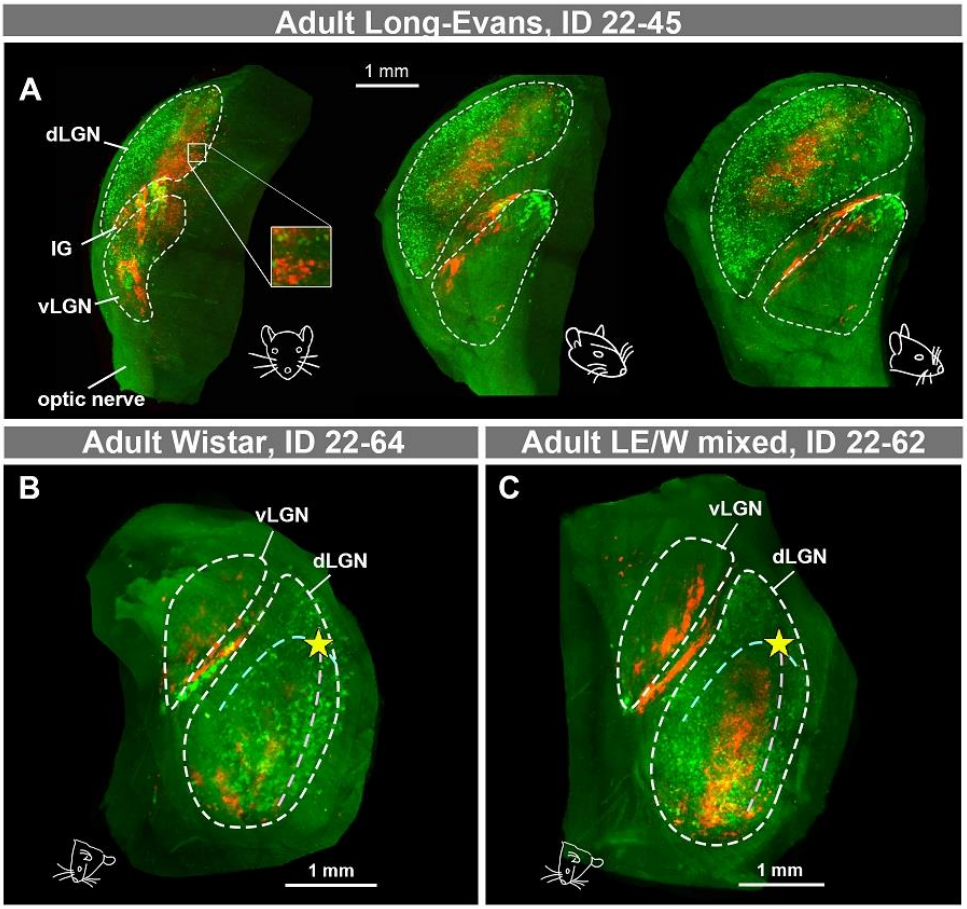
Topic: D.06. Vision

Support: NSFC 31872767
NSFC 32170992
NSFC U20A20221
China Brain Initiative 2021ZD0200401
Zhejiang Province Natural Science Foundation LR20C070002

Title: Three-dimensional topography of eye-specific domains in the lateral geniculate nucleus of pigmented and albino rats

Authors: *H. LI[1,2], Q. ZHOU[1], Y. CHEN[3], H. HU[3], L. GAO[3], T. TAKAHATA[1,2];
[1]Sch. of Med., Zhejiang Univ., Hangzhou, China; [2]Key Lab. for Biomed. Engin. of Ministry of Educ., Col. of Biomed. Engin. and Instrument Science, Zhejiang Univ., Hangzhou, China; [3]Key Lab. of Structural Biol. of Zhejiang Province, Sch. of Life Sciences, Westlake Univ., Hangzhou, China

Abstract: We recently revealed the presence of ocular dominance columns (ODCs) in the primary visual cortex (V1) of pigmented rats, but not in albino rats. Other studies have shown that the ipsilateral eye-domains of the dorsal lateral geniculate nucleus (dLGN) are segregated into patches in pigmented rats. Therefore, we were interested in the relationship between V1 ODCs and dLGN patches. To investigate how the dLGN patches are developed, how different they are between rat strains, and how much they are dependent on visual experience, we injected different tracers into the right and left eyes of rats and examined the labeling in the dLGN. Furthermore, we applied the tissue clearing technique to reveal the three-dimensional morphology of the dLGN. Our results showed that 1) The eye-specific segregation and the binocular zone organization in the dLGN are formed before the eyes open and do not change much afterwards. In a specific angle of the binocular zone of the dLGN, which we term ‘the retinotopic view’, the eye-specific patches can be seen similarly to ODCs of V1; 2) There are less ipsilateral domains in the dLGN of albino rats than in pigmented rats, although the ipsilateral patches are present even in albino rats. During development, the ipsilateral domains stay little in albinos, while they slightly increase in pigmented rats; 3) Eye-specific segregation in the dLGN is only moderately affected even when we imbalance the eye input during the precritical and critical period. Altogether, these anatomical results provide insights into the development of eye-specific geniculo-cortical network in rats regarding the basis of ODC formation.



Disclosures: **H. Li:** None. **Q. Zhou:** None. **Y. Chen:** None. **H. Hu:** None. **L. Gao:** None. **T. Takahata:** None.

Poster

PSTR149. Functional Organization of the Developing and Mature Visual System

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR149.02/Z26

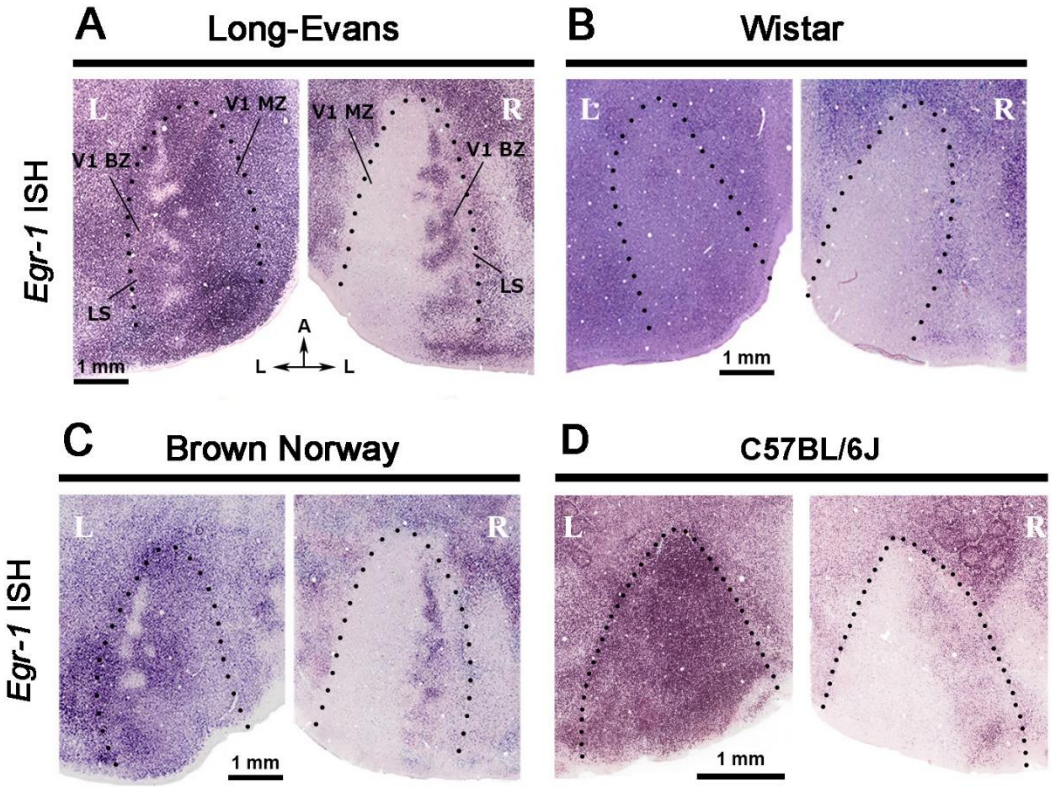
Topic: D.06. Vision

Support: NSFC 31872767
NSFC 32170992
NSFC U20A20221
China Brain Initiative 2021ZD0200401

Title: Characterization of ocular dominance columns in rodents.

Authors: *Q. ZHOU[1], H. LI[1,2], S. YAO[1,2], T. TAKAHATA[1,2];
[1]Interdisciplinary Inst. of Neurosci. and Technology, Sch. of Med., [2]Key Lab. for Biomed. Engin. of Ministry of Education, Col. of Biomed. Engineerin, Zhejiang Univ., Hangzhou, China

Abstract: Despite previous agreement of the absence of cortical columns in the rodent visual cortex, we have recently revealed a presence of ocular dominance columns (ODCs) in the primary visual cortex (V1) of adult Long-Evans rats. In this study, we deepened understanding of characteristics of rat ODCs. By examining the expression pattern of immediate-early gene (IEG) *Egr-1* mRNA in the V1 following monocular enucleation, we found that ODCs were conserved in Brown Norway rats, but completely absent in albino rats. Therefore, ODCs could be a structure generally present in pigmented wild rats, indicating their possible functional importance for survival of animals in wild environment. *Egr-1* mRNA expression pattern indicated that the ODCs of rats gradually get matured after eye-opening. At postnatal 5 weeks, the structure of ODCs is almost comparable to that of adult cases, and this developmental process is visual experience dependent. Monocular deprivation by eyelid suture during classical critical period strongly influenced size of ODCs, shifting ocular dominance from the deprived eye to the opened eye. Dark-rearing also abolished the normal development of ODCs. On the other hand, transneuronal anterograde tracer showed a presence of eye-dominant patchy innervation from the ipsilateral V1 even before eye-opening, suggesting the presence of visual activity-independent genetic components of developing ODCs. Intriguingly, pigmented C57BL/6J mice also showed minor clusters of ocular dominance neurons, which are probably prototype of ODCs. These results provide insights into how ODCs are conserved across different rodent species and strains, how visual experience-dependent and -independent components both contribute to develop ODCs during early postnatal stages, and indicate that rats and mice can be excellent models to study them.



Disclosures: **Q. Zhou:** None. **H. Li:** None. **S. Yao:** None. **T. Takahata:** None.

Poster

PSTR149. Functional Organization of the Developing and Mature Visual System

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR149.03/Z27

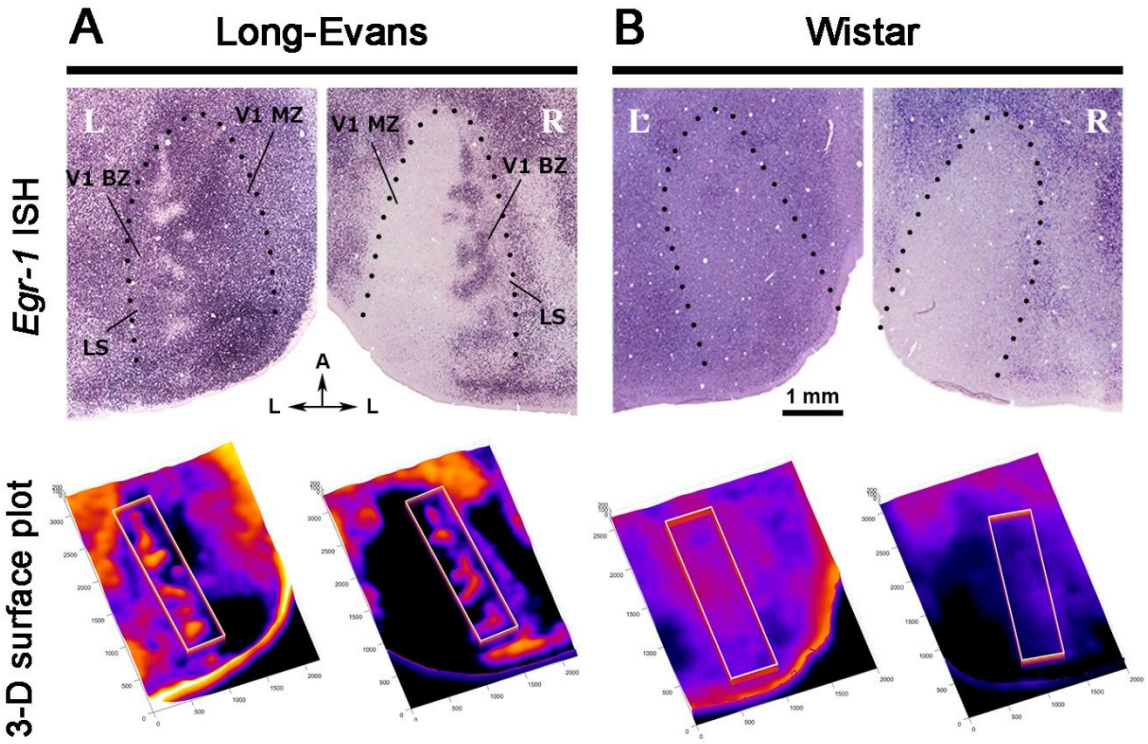
Topic: D.06. Vision

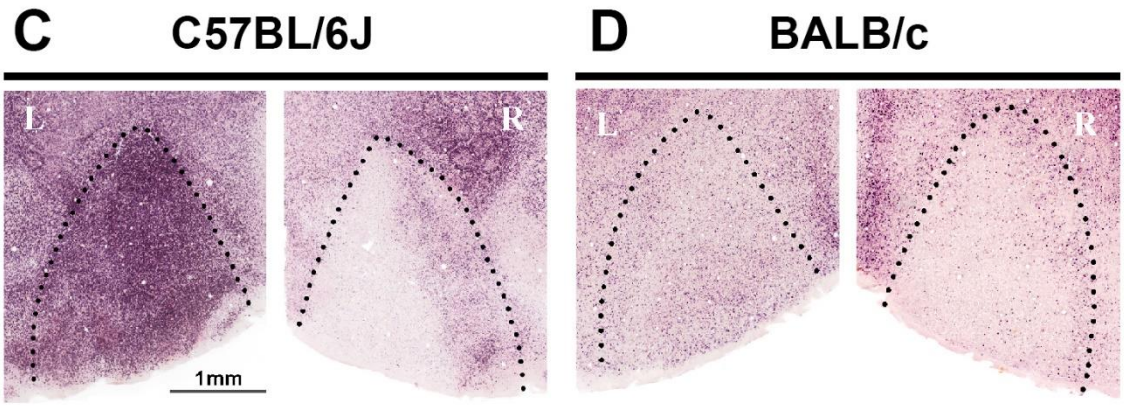
Support: NSFC 31872767
NSFC 32170992
NSFC U20A20221
China Brain Initiative 2021ZD0200401

Title: Development and plasticity of ocular dominance columns in rodents.

Authors: *T. TAKAHATA;
Zhejiang Univ., Zhejiang, China

Abstract: “Cortical columns” had been considered to specifically present in highly evolved brains, but not in rodents. We have revealed, however, the presence of ocular dominance columns (ODCs) in the primary visual cortex (V1) of Long-Evans rats. The expression patterns of immediate-early genes (IEGs) clearly depicted patchy territories of the right and left eye domains throughout layers in the binocular zones of V1 following monocular inactivation. These patches were completely absent in V1 of albino rats, but seen in Brown Norway rats as well, implicating broad conservation of ODCs in pigmented rat strains, including wild rats. The IEG patches were not observed at the time of eye-opening, but became clearer between postnatal day (P) 21 and P35. Dark-rearing and monocular deprivation treatments both abolished formation of the IEG patches. Interestingly, ODCs revealed by ipsilateral anterograde tracing of WGA-HRP appeared much earlier. This paradox suggests time-gap between development of geniculo-cortical afferents and visually-driven responses of cortical neurons, and perhaps the period of this gap was recognized as “critical period” for ocular dominance plasticity in previous studies. ODCs were not clearly observed in the pigmented mouse V1, but yet minor clusters of IEG expression were seen, possibly showing prototype of ODCs. Consequently, our finding elicits a paradigm shift on species conservation of cortical column and identity of plasticity during classical critical period for ocular dominance.





Disclosures: **T. Takahata:** None.

Poster

PSTR149. Functional Organization of the Developing and Mature Visual System

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR149.04/Z28

Topic: D.06. Vision

Support: NIH R01EY006821

Title: Sequential emergence of excitatory and inhibitory functional modular networks in developing visual cortex

Authors: *A. GRIBIZIS, R. SATTERFIELD, D. FITZPATRICK;
Max Planck Florida Inst. for Neurosci., Jupiter, FL

Abstract: In the primary visual cortex (V1) of many mammals, orientation preference is organized in a periodic pattern across cortical space known as orientation maps. Patterned spontaneous cortical activity immediate before eye opening in V1 resembles the modular topology of orientation maps, consistent with an experience-independent origin of this fundamental modular structure. However, the sequence of events that leads to the initial emergence of modular network structure early in development remains unclear.
Here we explore the onset of excitatory and inhibitory networks in the developing visual cortex of the tree shrew. The mature visual cortex of the tree shrew has long served as a model for studying the functional organization of circuits in V1 due to its well-defined modular representation of visual properties and resemblance to primate V1.
Using multi-color imaging to measure calcium signals in genetically targeted cell types, we have developed a chronic imaging preparation to visualize changes in patterns of V1 spontaneous activity over several days early in development. We find that the earliest patterns of both excitatory and inhibitory spontaneous activity are evident several days prior to eye opening have

the appearance of large, isolated single patches. Following these early coarse solitary patterns of activity, distinctly finer-scale modular network patterns of activity suddenly emerge, with coactivation of multiple patches extending millimeters across the cortical surface.
Ongoing studies using multiphoton functional imaging, laminar electrophysiological probes, and structural analyses of cell morphology are evaluating structural changes that could be developing concurrently with this rapid emergence of modular networks.

Disclosures: **A. Gribizis:** None. **R. Satterfield:** None. **D. Fitzpatrick:** None.

Poster

PSTR149. Functional Organization of the Developing and Mature Visual System

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR149.05/AA1

Topic: D.06. Vision

Support: IARPA Grant D16PC00003
IARPA Grant D16PC00004
IARPA Grant D16PC00005
NSF CAREER Grant IOS-1552868
NSF NeuroNex grant 1707400
National Institute of Mental Health and National Institute of Neurological Disorders And Stroke under Award Number U19MH114830
National Eye Institute award numbers R01 EY026927
Core Grant for Vision Research T32-EY-002520-37
NLM Training Program in Biomedical Informatics & Data Science (T15LM007093)

Title: Functional connectomics reveals general wiring rule in mouse visual cortex

Authors: *Z. DING[1,2], P. FAHEY[1,2], S. PAPADOPOULOS[1,2], E. Y. WANG[1,2], B. CELII[1,2], C. PAPADOPOULOS[1,2], A. KUNIN[4,1,2], A. CHANG[1,2], J. FU[1,2], Z. DING[1,2], S. S. PATEL[1,2], K. PONDER[1,2], T. MUHAMMAD[1,2], M. CONSORTIUM[3,5,6], E. FROUDARAKIS[7,8], F. H. SINZ[9,10], H. SEUNG[5], F. COLLMAN[6], N. DA COSTA[6], C. REID[6], E. Y. WALKER[11,12], X. S. PITKOW[1,2,13], J. REIMER[1,2], A. S. TOLIAS[1,2];
[1]Neurosci., [2]Ctr. for Neurosci. and Artificial Intelligence, [3]Baylor Col. of Med., Houston, TX; [4]Dept. of Mathematics, Creighton Univ., Omaha, NE; [5]Princeton Univ., Princeton, NJ; [6]Allen Inst. For Brain Sci., Seattle, WA; [7]Dept. of Basic Sciences, Fac. of Med., Univ. of Crete, Heraklion, Greece; [8]Inst. of Mol. Biol. and Biotech., Fndn. for Res. and Technol. Hellas, Heraklion, Greece; [9]Inst. for Bioinformatics and Med. Informatics, Univ. Tübingen, Tübingen, Germany; [10]Inst. for Computer Sci. and Campus Inst. Data Sci., Univ. Göttingen, Göttingen, Germany; [11]Dept. of Physiol. and Biophysics, [12]Computat. Neurosci. Ctr., Univ. of Washington, Seattle, WA; [13]Dept. of Electrical and Computer Engin., Rice Univ., Houston, TX

Abstract: To understand how the brain computes, it is important to unravel the relationship between circuit connectivity and function. Previous research has shown that excitatory neurons in layer 2/3 of the primary visual cortex of mice with similar response properties are more likely to form connections. However, technical challenges of combining synaptic connectivity and functional measurements have limited these studies to few, highly local connections. Utilizing the millimeter scale and nanometer resolution of the MICrONS dataset, we studied the connectivity-function relationship in excitatory neurons of the mouse visual cortex across interlaminar and interarea projections, assessing connection selectivity at the coarse axon trajectory and fine synaptic formation levels. A digital twin model of this mouse, that accurately predicted responses to arbitrary video stimuli, enabled a comprehensive characterization of the function of neurons. We found that neurons with highly correlated responses to natural videos tended to be connected with each other, not only within the same cortical area but also across multiple layers and visual areas, including feedforward and feedback connections.The digital twin model separated each neuron's tuning into a feature component (what the neuron responds to) and a spatial component (where the neuron's receptive field is located). We show that the feature, but not the spatial component, predicted which neurons were connected at the fine synaptic scale. Together, our results demonstrate the “like-to-like” connectivity rule generalizes to multiple connection types, and the rich MICrONS dataset is suitable to further refine a mechanistic understanding of circuit structure and function.

Disclosures: **Z. Ding:** None. **P. Fahey:** None. **S. Papadopoulos:** None. **E.Y. Wang:** None. **B. Celii:** None. **C. Papadopoulos:** None. **A. Kunin:** None. **A. Chang:** None. **J. Fu:** None. **Z. Ding:** None. **S.S. Patel:** None. **K. Ponder:** None. **T. Muhammad:** None. **M. Consortium:** None. **E. Froudarakis:** None. **F.H. Sinz:** None. **H. Seung:** None. **F. Collman:** None. **N. da Costa:** None. **C. Reid:** None. **E.Y. Walker:** None. **X.S. Pitkow:** None. **J. Reimer:** None. **A.S. Tolias:** None.

Poster

PSTR149. Functional Organization of the Developing and Mature Visual System

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR149.06/AA2

Topic: D.06. Vision

Support: NIH Grant R01EB029813
NIH Grant R01NS122742
NIH Grant U24NS124001

Title: Exploring neuronal types, connectivity, and synaptic kinetics in a data-driven model of mouse V1

Authors: *D. HAUFLER, S. ITO, K. DAI, X.-P. LIU, N. REN, A. ARKHIPOV;
Allen Inst., Seattle, WA

Abstract: A comprehensive understanding of cognitive activity in the neocortex necessitates an in-depth accounting of neuronal and synaptic heterogeneity. To gain insight into the respective contributions of neuronal cell types to visual processing, we have developed a data-driven model of mouse primary visual cortex (V1) utilizing the latest large-scale data resources from the Allen Institute characterizing mouse V1. Building on our group's established V1 model, we incorporate results from the Synaptic Physiology dataset [https://brain-map.org/explore/connectivity/synaptic-physiology], which utilizes multi-patch electrophysiological probing of local connectivity in transgenic mouse lines, and the IARPA MICrONS dataset [https://www.microns-explorer.org/], consisting of a ~1 cubic mm of digitally reconstructed tissue obtained via electron microscopy. The model upgrades we implemented refine connection probabilities, connection weight distributions, synaptic kinetics, and heterogeneity in synapse numbers across neurons which replace aggregated parameter values or approximations from the literature. Simulations of the new model reveal various distinctions from its predecessor including stability prior to optimization and better expression of physiological rhythms. Augmentations of the model also allow us to characterize the impact of specific structural properties on visual processing: We find a nonlinear dependence of orientation and direction tuning of neurons on rules governing assignment of connections, weights, and spatial distribution of inputs whereby tuning properties are preserved when these rules are removed individually but not in combination. We expect that this model will provide a valuable platform for the scientific community to further explore and decode the structure and function of cortical circuits.

Disclosures: **D. Haufler:** None. **S. Ito:** None. **K. Dai:** None. **X. Liu:** None. **N. Ren:** None. **A. Arkhipov:** None.

Poster

PSTR149. Functional Organization of the Developing and Mature Visual System

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR149.07/AA3

Topic: D.06. Vision

Support: NIH Grant EY022122

Title: Co-organization of responses to direction, speed, and temporal frequency in ferret visual cortex

Authors: *V. SUÁREZ CASANOVA[1,2], N. LASKY-NIELSON[2], X. CHENG[2], R. RODRIGUEZ[1], J. TOUBOUL[3], J. RIBOT[4], S. D. VAN HOOSER[2];
[1]Biol., Brandeis Univ. Grad. Neurosci. Program, Waltham, MA; [2]Biol., [3]Mathematics, Brandeis Univ., Waltham, MA; [4]Collège de France-CNRS, Col. de France, Paris, France

Abstract: To perceive a moving object in an animal’s visual environment, it is necessary for the visual system to interpret the direction and speed of the objection as well as its spatial properties:

orientations and spatial frequencies. Decoding the spatial characteristics of a stimulus is well understood: tuning in primary visual cortex (V1) for specific properties - stimulus orientation and spatial frequency (SF) measured in cycles per degree of visual angle - does not change when other parameters of stimulus are modulated. But, how the visual cortex might code the temporal properties - direction, temporal frequency (TF), and speed - is incompletely understood. It has been known for some time that a subpopulation of neurons alter their direction preferences as TF is altered (Moore et al. 2004), and recent unpublished data from author Ribot shows that this is true for large portions of the carnivore visual cortex. Therefore, a one-dimensional population read-out of stimulus direction is not possible for this population of cells. We report progress on a study of the functional organization underlying stimulus temporal processing, namely the relationship between SF, TF and direction using acute in-vivo electrophysiology and two-photon calcium imaging in the ferret primary visual cortex. We hypothesize that the activity of V1 neurons contains the information necessary to decode stimulus direction, speed, and temporal frequency in a non-linear manner. We will examine whether organization of the temporal tuning properties is maintained across visual cortical layers, or if temporal tuning characteristics, speed tuning, emerge across the layers. Our preliminary results suggest that speed-sensitive cells are clustered across the surface of cortex. These results will elucidate the structure of the receptive field of these cells across layers and may provide answers to functional architecture underlying these properties and whether they share similar computing principles.

Disclosures: V. Suárez Casanova: None. **N. Lasky-Nielson:** None. **X. Cheng:** None. **R. Rodriguez:** None. **J. Touboul:** None. **J. Ribot:** None. **S.D. Van Hooser:** None.

Poster

PSTR149. Functional Organization of the Developing and Mature Visual System

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR149.08/AA4

Topic: D.06. Vision

Support: Dfg Emmy Noether: VE 938/2-1

Title: Context-dependent interareal synchronization across mouse visual cortex

Authors: *C. RAMANATHAN, D. ERIKSSON, J. VEIT;
Inst. of Physiology, Dept. 1, Albert Ludwigs Univ. of Freiburg, Freiburg im Bresigau, Germany

Abstract: Most brain functions rely on constant interaction between specialized brain areas. In primates, it has been suggested that synchronized oscillations in distinct frequency bands could serve to route feedforward and feedback signals across hierarchically organized visual areas. However, the underlying circuit mechanisms are still unknown. We utilized visually induced oscillations in our own recordings, as well as the Allen Institute Neuropixels visual coding data set to study inter-areal interactions across mouse visual cortical areas.In particular, we are investigating the synchronization of distinct visually induced rhythms across multiple visual

cortical areas. Two prominent visually induced gamma rhythms have been previously reported in mice - (i) a context-dependent visually induced low gamma rhythm (~30Hz) in V1 and (ii) a narrowband, luminance-dependent high gamma rhythm (~60Hz) in V1 and in higher visual areas. Our analysis of the Allen Institute data set reveals that the visually induced low-frequency (~30Hz) gamma oscillations is also present, at varying strengths, across cortical layers and across the entire visual cortical hierarchy. To better understand the visual stimulus dependence, the laminar profile, and the coherence of this rhythm across different areas, we performed simultaneous multi-areal extracellular recordings in retinotopically matched locations in V1 and LM (homolog of primate V2) of awake, head-fixed mice using Neuropixels probes.The low- and high-frequency gamma rhythms exhibit distinct stimulus dependence and laminar profiles in V1 and LM. Additionally, the interareal synchronization patterns of the two different rhythms between V1 and LM are distinct and dependent on stimulus, layer and retinotopy. We are currently further investigating the circuit mechanisms, as well as the directionality of these rhythms both analytically - by performing Granger causality analysis, as well as experimentally - by optogenetic perturbation of retrogradely labeled feedforward and feedback projection neurons.

Disclosures: **C. Ramanathan:** None. **D. Eriksson:** None. **J. Veit:** None.

Poster

PSTR149. Functional Organization of the Developing and Mature Visual System

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR149.09/AA5

Topic: D.06. Vision

Support: NIH Grant EY023037

Title: Electrophysiological and calcium imaging signatures of visual recognition memory across all layers of mouse V1.

Authors: *D. P. MONTGOMERY[1,2], D. J. HAYDEN[3], M. F. BEAR[1,2];
[1]Brain and Cognitive Sci., [2]Picower Inst. for Learning and Memory, MIT, Cambridge, MA;
[3]Kavli Inst. for Systems Neurosci., Trondheim, Norway

Abstract: Recognition of familiar visual stimuli enables animals, from mice to humans, to efficiently navigate their world and detect novel, potentially behaviorally salient stimuli. One robust neurophysiological report of visual recognition memory is stimulus-selective response plasticity (SRP) in mouse visual cortex (V1). SRP can be elicited in mice that passively view the same visual stimulus across multiple days. SRP is most readily observed as a pronounced increase in the magnitude of visual evoked potentials (VEPs) recorded from V1 layer (L) 4 in response to phase-reversing grating stimuli. However, recent evidence points to the involvement of circuitry beyond L4 in the expression of SRP. We have therefore used multiple approaches to investigate how SRP is expressed across all cortical layers in order to gain a clearer picture of

how the cortical microcircuit supports this form of plasticity. Current-source density (CSD) analysis of the VEP depth profile shows augmentation of short latency current sinks in layers 3, 4 and 6 in response to phase reversals of familiar stimuli. Multi-unit recordings demonstrate that increased peak firing occurs in response to phase reversals of familiar stimuli across all layers, but that activity between phase reversals is suppressed. 2-photon calcium imaging reveals an average decrease in activity during familiar stimulus viewing in the somata of principal cells in L2/3 and L4, but no detectable change in deeper layers. However, imaging of the apical dendrites of L5 pyramidal cells reveals enhanced activity as animals view the familiar visual stimuli, whereas the apical dendrites of L6 corticothalamic neurons show diminished activity. Together, these data reveal important aspects of the underlying phenomenology of SRP and highlight important differences in what can be revealed by various measures of cortical activity in mice.

Disclosures: D.P. Montgomery: None. **D.J. Hayden:** None. **M.F. Bear:** None.

Poster

PSTR149. Functional Organization of the Developing and Mature Visual System

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR149.10/AA6

Topic: D.06. Vision

Support: NIH Grant MH099045
NIH Grant MH121841
NIH Grant EY022951
NIH Grant MH113852
Kavli Institute of Neuroscience
Simons Foundation SFARI Research Grant
Swebilius Foundation award
Support from the Ludwig Foundation

Title: Learning dependent reconfiguration of cortical circuits underlying visual fear conditioning

Authors: *A. H. MOBERLY, M. J. HIGLEY, J. A. CARDIN;
Neurosci., Yale Univ., New Haven, CT

Abstract: Forming associations between sensory cues and salient outcomes is fundamental for normal behavior and requires coordinated activity across distributed brain circuits. A large body of research has described the molecular mechanisms of plasticity at cellular and synaptic scales that are necessary for learning and memory. However, the circuit-level dynamics and plasticity supporting associative learning remain unresolved. Several studies have demonstrated a role for neocortical circuits in processing sensory information as a component of conditioned behavior. However, the extent to which learning drives plasticity of cortical representations and network dynamics in primary and secondary cortical areas is still unclear. In particular, the neural mechanisms that support the establishment of links between visual stimuli and emotional valence

that influence behavior are essentially unknown. Here, we developed a novel visually-cued task in which head-fixed mice learn to associate a 5 second visual stimulus (either a contrast-modulated patch of filtered noise or a drifting grating) with a mild foot-shock, using lick suppression as the behavioral readout of conditioning. We find that visually-cued lick suppression is contrast dependent and impaired by acute inactivation of visual cortex. Furthermore, mice readily learn a discrimination version of the task, exhibiting conditioned lick suppression to a stimulus paired with the shock and no suppression to an unpaired stimulus of orthogonal orientation. Using this behavioral framework, we monitored neuronal activity across the neocortex before and after conditioning using widefield mesoscopic calcium imaging. After learning, we observed a broad increase in visual cue-evoked activity across the cortex, with the greatest enhancement in medial higher-order visual areas. Cellular resolution 2-photon imaging in visual cortex revealed that conditioning also drives an increase in the evoked activity of vasoactive intestinal peptide (VIP) expressing interneurons. Given the potential role of these cells in behavioral state-dependent disinhibition of cortical circuits, these results suggest a similar mechanism may contribute to cortical plasticity during fear conditioning. Finally, viral tracing demonstrated that these regions are reciprocally coupled with subregions of the amygdala, suggesting a circuit mechanism by which fear conditioning strengthens interactions between sensory and limbic areas. Overall, our results demonstrate a novel paradigm in which learning drives plasticity of sensory representations in the visual cortex.

Disclosures: **A.H. Moberly:** None. **M.J. Higley:** None. **J.A. Cardin:** None.

Poster

PSTR149. Functional Organization of the Developing and Mature Visual System

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR149.11/AA7

Topic: D.06. Vision

Support: NIH National Eye Institute R01EY030893-01
Bundesministerium für Bildung und Forschung 01GQ2002
Whitehall Foundation (2018-05-57)
NIH National Institute of Mental Health MH115688 T32
National Science Foundation IIS-2011542

Title: Selective amplification of recurrent subnetworks in the developing visual cortex

Authors: *H. N. MULHOLLAND[1], S. TRÄGENAP[2], M. KASCHUBE[2], G. B. SMITH[1,3]; [1]Neurosci., Univ. of Minnesota-Twin Cities, Minneapolis, MN; [2]Frankfurt Inst. for Advanced Studies, Frankfurt am Main, Germany; [3]Optical Imaging and Brain Sci. Med. Discovery Team, Univ. of Minnesota, Minneapolis, MN

Abstract: Prior to visual experience, spontaneous activity in the primary visual cortex forms low-dimensional, modular activity patterns that reveal functional networks with millimeter-scale

correlations. Computational modeling demonstrates that heterogeneous local connections can produce long-range correlations, creating preferentially interacting recurrent subnetworks. These models predict that stimulus inputs that align with the structure of these endogenous subnetworks would be recurrently amplified, leading to more reliable evoked responses and constraining the potential outputs of the network. Using spatially structured optogenetic stimulation and *in* vivo calcium imaging in young ferret visual cortex before eye opening, we tested whether the alignment of optogenetic stimuli with the architecture of developing cortical networks determines the structure of the evoked response. We found that optogenetic stimuli based on endogenous patterns of spontaneous activity evoked responses that had greater reliability from trial-to-trial and were more similar to their input pattern compared to activity evoked by artificial stimuli that had similar spatial frequencies, but were poorly aligned with spontaneous activity. The degree of reliability and stimulus similarity was predicted by the amount of overlap the stimulus input pattern had with the principal components of spontaneous activity. Further, artificial stimuli did not create new activity patterns, but instead evoked activity that resided within the space spanned by endogenous network activity, indicating a network transformation that constrains possible outputs. Together, these results indicate that already before eye opening, early cortical activity is organized into preferentially connected millimeter-scale subnetworks that recurrently amplify input drives that align with the endogenous network architecture, providing insight into how reliable responses to sensory input might be built over the course of development.

Disclosures: H.N. Mulholland: None. **S. Trägenap:** None. **M. Kaschube:** None. **G.B. Smith:** None.

Poster

PSTR149. Functional Organization of the Developing and Mature Visual System

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR149.12/AA8

Topic: D.06. Vision

Support: 5R01EY011488

Title: Chronic, three-dimensional, extracellular recordings in developing ferrets reveal the sequence of functional changes in V1 following eye opening.

Authors: *A. A. LEMPEL, C. TEPOHL, D. FITZPATRICK;
Max Planck Inst. Florida, Jupiter, FL

Abstract: When an animal first opens its eyes, it is exposed to novel visual input containing edges and complex shapes. Within days of this developmentally critical event, V1 undergoes a myriad of functional changes. One of these transformations is the emergence of a modular representation of edge orientation that is columnar (consistent across cortical layers) and binocularly matched (consistent when stimuli are presented to either or both eyes). Still, the

sequence of changes that manifest across the V1 circuit to build a columnar and binocularly matched representation remains unclear. To study the emergence and maturation of functional properties in V1, we employed novel tools and methods for chronic, three-dimensional, extracellular recordings in developing ferrets around the time of natural eye opening (fifth week post-natal). Stimuli-evoked responses could be recorded from neurons in layers 2/3 (L2/3) and layer 4 (L4) of multiple functional columns right after eye opening and every 24-48 hours during the first week of visual experience. The recordings exhibited high stability, which allows for analyzing the developmental trajectory of functional columns and their response properties. In our first experiments, we measured responses to monocular and binocular stimulation with drifting gratings. Preliminary data on binocular responses at the time of eye-opening show a modular structure of neuronal activity correlation in L2/3 across functional domains. In contrast, responses in L4 did not display such a structure. 2-3 days later, a columnar representation of orientation emerged through correlated responses between layers and functional domains. Surprisingly, the developmental trajectory of these responses suggests that modular patterns of correlated activity in L2/3 instruct functional changes in L4. Preliminary data on monocular responses confirm that the process of binocular alignment continues to progress following the initial emergence of a columnar representation of orientation. Moreover, this process involved multiple transformations that differ between contralateral and ipsilateral stimulation. In particular, contralateral responses display stronger selectivity at the time of eye opening that remain more consistent over development compared to ipsilateral responses, suggesting the former instructs changes in the latter. These novel experiments not only provide a specific sequence and timeline for changes in response properties driven by early visual experience but can also define the developmental trajectories and relationship of such changes across different components of the V1 circuit.

Disclosures: **A.A. Lempel:** None. **C. Tepohl:** None. **D. Fitzpatrick:** None.

Poster

PSTR149. Functional Organization of the Developing and Mature Visual System

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR149.13/AA9

Topic: D.06. Vision

Support: JSPS KAKENHI Grant Number JP19K12743

Title: A mathematical study of competitive generation and elimination of dendritic spines in the postnatal development of the mammalian visual cortex

Authors: *M. MIYASHITA, S. TANAKA;
Natl. Inst. of Technol., Numazu, Shizuoka, Japan

Abstract: In the developing brain, neural circuits are elaborated by the activity-dependent rewiring of synaptic connections. Our earlier self-organization model built on the assumption

that presynaptic axons contact competitively with single dendritic spines successfully reproduced spatiotemporal receptive fields and orientation-direction maps. However, microscopic observations have proven that synaptic rewiring occurs more often by morphological changes in dendritic spines than by those in presynaptic terminals. It has also been reported that most synaptic contacts are *en passant* types. This fact is contradictory to our assumption that each axon terminal selects a proper dendritic spine in an activity-dependent manner. On the other hand, it has been proposed that individual spines interact with each other by the diffusion of Rho-GTPases in the dendrites. In the present study, we attempt to rebuild the mathematical model, taking into account the following experimental observations: (1) Intracellular Ca2+ concentration increases by Ca2+ influx through NMDAR and is enhanced by the occurrence of backpropagating action potential, (2) BDNF is released in a Ca2+ concentration-dependent manner, (3) activity-dependent release of tPA converts plasminogen to plasmin, (4) plasmin cleaves proBDNF to matured BDNF and BDNF propeptide, (5) binding matured BDNF to TrkB activates Cdc42, (6) binding BDNF propeptide or proBDNF to p75NTR activates RhoA, (7) activated RhoA diffusing in the dendrite and contributes to actin depolymerization leading to the shrinkage of adjacent dendritic spines, and 8) activated Cdc42 promotes actin polymerization resulting in the elongation of spines. The simulations based on the new model reproduced simple-cell-like receptive fields and orientation-direction joint maps. Furthermore, we obtained a characteristic profile of the total number of synapses that shows early increase and late decrease during the critical period.

Disclosures: **M. Miyashita:** None. **S. Tanaka:** None.

Poster

PSTR149. Functional Organization of the Developing and Mature Visual System

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR149.14/AA10

Topic: D.06. Vision

Support: NEI
NINDS
Revson Foundation

Title: Inhibition enhances encoding precision of neuronal ensembles in mouse visual cortex

Authors: *J. PEREZ-ORTEGA, A. AKROUH, R. YUSTE;
Columbia Univ., New York, NY

Abstract: Neuronal ensembles, coactive groups of neurons, are associated with motor, sensory, and cognitive functions. However, the mechanisms underlying their information encoding remain unclear. In this study, we employed two-photon multiplane calcium imaging to investigate the functional properties of ensembles in the primary visual cortex of awake mice during visual stimulation. Using a novel unsupervised algorithm, we identified significant

coactivation patterns to extract ensembles. Surprisingly, ensembles exhibited higher orientation selectivity and narrower bandwidth compared to individual neurons. Ensembles also outperformed tuned neurons in decoding visual stimuli, displaying superior sensitivity and specificity. Furthermore, our characterization of ensemble and non-ensemble neurons revealed that the activation of an ensemble led to the inhibition of a distinct set of neurons, referred to as "offsembles". Interestingly, offsembles alone accurately predicted the orientation of visual stimuli, but the combined activity of ensemble and offsemble neurons yielded an enhanced precision in stimulus encoding. These findings underscore the significance of the ensemble-offsemble partnership as an emergent and distributed neural code within cortical circuits, encoding information with enhanced precision.

Disclosures: J. Perez-Ortega: None. **A. Akrouh:** None. **R. Yuste:** None.

Poster

PSTR149. Functional Organization of the Developing and Mature Visual System

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR149.15/AA11

Topic: D.06. Vision

Support: NIH 1R01EY031059
NSF Grant 1822598

Title: Dimensionality Varies Across Manifold Representations of Mouse Visual Neurons

Authors: *L. DYBALLA[1], A. M. RUDZITE[3], M. HOSEINI[4], M. THAPA[5], M. P. STRYKER[4], G. D. FIELD[5], S. W. ZUCKER[2];
[2]Computer Sci., [1]Yale Univ., New Haven, CT; [3]Neurobio., Duke Univ., Durham, NC; [4]Ctr. for Integrative Neurosci, Dept Physiol, Univ. of California, San Francisco, San Francisco, CA; [5]Stein Eye Institute, Dept Ophthalmology, UCLA, Los Angeles, CA

Abstract: Understanding how networks of neurons represent visual features and how these relate across different areas remains a challenge. We have addressed this problem using a neural encoding manifold (Dyballa et al, 2023, bioRxiv). Each point on the manifold is a neuron, and neurons are organized by their responses to an ensemble of stimuli (artificial gratings and naturalistic flows). Neurons nearby on the manifold respond similarly in time to similar stimulus components, and are thus likely to be involved in network activity. Stimuli subtending ~60^o x 40^o were presented for 1.25 sec separated by 0.75 sec mid-gray screens. Responses were recorded ex-vivo from 1149 neurons in 3 retinas using a 519-channel multielectrode array. Recordings from 658 neurons in the primary visual cortex of 12 awake, behaving mice were made with a 128-channel silicon probe. The manifold was inferred using diffusion maps with an adaptive kernel.
The intrinsic dimensionality of the manifold indicates how many 'degrees of freedom' (DOF) there are in each neuron's behavior, including variation in both stimulus features (e.g., contrast or

orientation selectivity) and dynamics (e.g., transient vs. sustained). The retinal manifold topology is highly clustered while the cortical manifold is continuous. We now report that the estimated intrinsic dimensionality of the cortical manifold (>5) is about double that of the retinal manifold (~3) for our stimulus ensemble (Dyballa et al., 2018, PNAS). Low dimension might be expected for the retina, where clusters of RGCs correspond to a mosaic of distinct types, but the situation is richer for cortex. One DOF indicates a competition between low-frequency spatial gratings and all other stimuli, and another indicates how orientation tuning for high frequency gratings is sharpened in concert with oriented flows. Surprisingly, a third DOF indicates a preference for low frequency spatial gratings in concert with random dot flows, but in competition with oriented flows.
Taken together, these assessments of the neural encoding manifold are beginning to unravel the myriad functional implications of cortical recurrent circuitry, when compared against retina's largely feedforward organization.

Disclosures: **L. Dyballa:** None. **A.M. Rudzite:** None. **M. Hoseini:** None. **M. Thapa:** None. **M.P. Stryker:** None. **G.D. Field:** None. **S.W. Zucker:** None.

Poster

PSTR149. Functional Organization of the Developing and Mature Visual System

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR149.16/AA12

Topic: D.06. Vision

Support: NSF IIS-2113197
NIH Intramural Program

Title: Biologically constrained deep neural networks to parse visual computations being performed in the primary visual cortex

Authors: *M. JACOBSEN[1], F. BARTSCH[1], B. R. CONWAY[2], J. YATES[3], D. BUTTS[1]; [1]Univ. of Maryland, Col. Park Neurosci. and Cognitive Sci. Program, College Park, MD; [2]Natl. Inst. of Hlth., Bethesda, MD; [3]UC Berkeley, Oakland, CA

Abstract: The transformations performed by the visual system from the raw spatiotemporal pattern of light on the retina into our visual perception are the result of computations performed by a massive number of neurons across the visual cortex. Our understanding of how these computations are implemented by visual circuits is still limited by our ability to model the complex nonlinearities present in the system. On the one hand, understanding visual processing performed by neurons has been well characterized by simple visual stimuli, but these descriptions do not generalize to complex stimuli. On the other hand, recent advances applying convolutional deep neural networks (CNNs) to accurately predict responses of neurons in complex stimulus contexts thus far have been "black boxes" in terms of relating their internal computations to those performed by neurons and circuits.

Here, we present a biologically constrained CNN that can be fit to large-scale multi-electrode recordings of neurons in the primary visual cortex (V1). Our experiments used tailored correlated noise stimuli presented to an awake fixating macaque to strongly drive V1 nonlinear processing while avoiding much of the complexities of natural stimuli. To describe this data, we fit a CNN with several important structural elements that constrain its internal representations to be "interpretable" and more directly related to the neural data. These structural elements of the CNN include (1) a bottleneck at the first stage of processing that limits spatiotemporal processing to a small number of units with temporal and spatial scales of lateral geniculate nucleus (LGN) neurons; (2) applying Dale's Principle to constrain units within the CNN to be excitatory or inhibitory; and (3) temporal delays within the layers of the CNN to capture short-time scale cortical dynamics
After fitting this model to our recorded datasets, we found that the resulting structure of computation with the CNNs exhibited many known properties of circuits within the primate visual system. For example, the CNN's first layer resembled the spatiotemporal processing of LGN cells, including ON and OFF channels with magnocellular- and parvocellular-like properties. The second CNN layer contained Gabor-like units with PUSH-PULL connectivity, i.e., complementary patterns of ON- and OFF- excitation and inhibition. Finally, the deeper layers contained selectivity to increasingly large spatial and temporal scales, with response properties resembling extra-classical surrounds. Our biologically constrained CNN thus provides a foundation to understand how computation is structured across neurons in the visual cortex.

Disclosures: M. Jacobsen: None. **F. Bartsch:** None. **B.R. Conway:** None. **J. Yates:** None. **D. Butts:** None.

Poster

PSTR149. Functional Organization of the Developing and Mature Visual System

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR149.17/AA13

Topic: D.06. Vision

Support: NIH Grant R01EB031765
NIH Grant R01MH128177

Title: Decoding the visual orientation in cats' primary visual cortex with 9.4 Tesla laminar fMRI

Authors: *S. CHO[1], J. ZIMMERMANN[2];
[1]Radiology, Univ. of Minnesota, Saint Paul, MN; [2]Neurosci., Univ. of Minnesota, Minneapolis, MN

Abstract: Understanding how visual information is encoded and represented as neuronal population code has been a central question in functional magnetic resonance imaging (fMRI). Studies investigated the hemodynamic responses in the visual cortex, in which visual inputs evoke spatially distributed activation patterns across multiple voxels. Multi-voxel pattern

classification is the analysis technique in fMRI to decode the pattern feature and infer the visual inputs from the pattern reversely. However, the typical spatial resolution in fMRI decoding is insufficient to resolve the differential activations across cortical layers; therefore, it is unclear how voxels in different cortical layers distinctively contribute to decoding performance. Our study examined the accuracy of visual orientation decoding in voxels of different cortical layers (1 to 6). We employed the high-resolution layer fMRI (250 μm isotropic voxel in 9.4 Tesla fMR) to measure the cerebral blood volume changes responding to the four orientation gratings (0°, 45°, 90°, and 135°) in two cats' primary visual cortex. We then separately applied multi-voxel pattern classification for voxels of each cortical depth, calculating the cortical-depth-dependent orientation decoding accuracy. The results showed that, on average, the algorithm yielded 65.87% accuracy (chance level: 25%) in all layers. The highest accuracy was found in layers 1 voxels and 2/3 (85.12%), and the lowest was found in layer 4 voxels (50.92%). At all cortical depth points, the accuracy was greater than chance (25%); therefore, information for discerning orientations would remain in all layers, yet orientation-specific pattern features might appear in the upper layers (layers 1 to 3). In contrast, the accuracy in layer 4 was lower, implicating less discernable patterns among orientation conditions. These results would be relevant to findings of electrophysiology results that the neurons in cortical layer 2/3 exhibited more tuned responses to a specific orientation than those in layer 4, where massive projections deliver the orientation-non-specific visual input from the lateral geniculate nuclei. We conclude that fMRI decoding for visual orientation varies on the depth of region-of-interests, suggesting the cortical layer-specific multi-voxel pattern classification could achieve better decoding results by mainly focusing on the upper layers. Broadly, the approach will help further decipher the neural population codes for complex visual input and the input of other sensory modalities (i.e., auditory and motor) as it's expanded application.

Disclosures: S. Cho: None. **J. Zimmermann:** None.

Poster

PSTR149. Functional Organization of the Developing and Mature Visual System

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR149.18/AA14

Topic: D.06. Vision

Support: NSERC RGPIN-2018-06506
NSERC Postgraduate Scholarships – Doctoral (PGS D)

Title: Cortical cross-modal recruitment and functional connectivity in blind mice

Authors: *G. LALIBERTE[1], D. BOIRE[2];
[1]Anat., UQTR, Trois-Rivières, QC, Canada; [2]Anat., UQTR, Trois-Rivieres, QC, Canada

Abstract: Loss of vision results in cross-modal recruitment of the visual network by the remaining sensory modalities. The onset and duration of visual deprivation are crucial, as the

maturation of the visual pathways is influenced by spontaneous retinal waves (E16-P2) and visual afferent information during the critical period (P20-P35). In this study, we investigate cortical functional connectivity (FC) in sighted mice (n=14 C57Bl6 (C57), n=5 ZRDBA (Zr)) and in visually deprived mice by neonatal binocular enucleation (n=14 C57, n=3 Zr) and congenital anophthalmia (n=5 Zr) using mesoscopic calcium imaging with GCaMP6s reporters. The reporter is expressed in all neurons (hSyn; C57 and Zr) or specific neuronal populations (glutamatergic (CaMKIIa) or GABAergic (mDLX) neurons in C57). Cross-modal cortical activation involves strengthening existing neural pathways through long-term potentiation and adjusting excitatory/inhibitory balance, rather than forming new connections between sensory modalities. Considering the distinct feedback (FB) received by higher visual areas (HVAs), we hypothesize that these FB projections transition from modulators to drivers, enhancing cross-modal integration across visual cortices. In blind mice, auditory stimulation elicited responses in visual cortical areas, supporting cross-modal recruitment. Reduced interhemispheric FC was observed in V1, but not in spared sensory cortices (auditory (AU) and barrel field of S1). Cortical FC was similar between sighted C57 and Zr mice. However, FC between dorsal and ventral visual cortices was reduced while remaining largely similar within each stream both enucleated mice of these strains. Although, FC between lateral HVAs and AU was similar in sighted and enucleated C57 mice, it was stronger in enucleated and anophthalmia Zr than in intact sighted Zr mice. FC between V1 and S1 (limbs and trunk portion) increased following neonatal enucleation in both C57 and Zr mice, while increased FC was observed between somatosensory and motor areas in the in anophthalmia Zr mice. These observations suggest strain differences in their reaction to vision deprivation. These results indicate differential effect of enucleation and anophthalmia, and also strain related differences of cross-modal plasticity. Similar to humans, spontaneous retinal waves during subcortical visual pathways maturation, without visual evoked stimulation during the critical period, seem to have a greater effect on FC disruption in visual cortices than the absence of visual maturation in the occipital cortices.

Disclosures: **G. Laliberte:** None. **D. Boire:** None.

Poster

PSTR149. Functional Organization of the Developing and Mature Visual System

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR149.19/AA15

Topic: D.06. Vision

Support: FRQS - 312876
NSERC
Quebec bio-imaging network
CIRCA

Title: Longitudinal monitoring of the spontaneous activity of the dorsal cortex in late blind mice using widefield calcium imaging

Authors: *I. DJEROUROU, M. PTITO, M. VANNI;
École d'Optométrie, Univ. de Montréal, Montréal, QC, Canada

Abstract: The loss of visual inputs has major anatomical and functional effects on brain organization. In humans, the cross-modal plasticity that follows vision loss has been well characterized. However, the difficulty to recruit blind participants along with the heterogeneity of groups makes it difficult to study the development of cross-modal plasticity over time and in terms of the sensory experience of the individuals. Moreover, while most of the studies on blindness focus on early vision loss, few studies investigate the effect of vision loss at the adult stage on the brain, which represent the clinical situation of most blind people in the world.
The mouse is a popular model in neuroscience that has been used to study the effect of vision loss on the brain mostly using anatomical investigations. It has been found that it takes around 7 weeks after monocular enucleation to observe an activation of the monocular zone after stimulation of the whiskers. However, it is now relatively easy to express fluorescent calcium indicators to measure the neuronal activity of specific neuronal populations in awake mice over several weeks/months.
The brain expresses rich and dynamic activity patterns when the animal is not engaged in any specific task and in the absence of any external sensory stimuli. This “resting state” has been used to characterize the functional connectivity between brain regions. In the present study, we seek to explore how binocular enucleation in the adult mouse affects the spontaneous activity pattern of the dorsal cortex over time using calcium imaging.
We have implanted a chronic imaging window on 12 thy1-jrGECO1a mice to perform widefield calcium imaging on the dorsal cortex. Mice were group housed to maximize the multisensory experience and ultimately the cross-modal plasticity. They were habituated to be head-fixed on a running wheel under the imaging system. To capture the red fluorescence from jrGECO1a, the dorsal cortex was illuminated with a green LED (565/12nm), and the fluorescence, filtered at 618/50nm, was captured by a camera (CMOS). Retinotopic mapping with moving checkerboard bars on a monitor was performed to get the retinotopic maps and determine the position of visual areas of all mice. After 3 baseline sessions of spontaneous activity (10min), 6 mice were bilaterally enucleated while the 6 others remain sighted as a control. Imaging sessions were then conducted weekly.
Compared to previous investigations focusing on early blind mice, this study exploits the benefit of longitudinal monitoring to map the changes in connections induced by the loss of vision after the period of development.

Disclosures: I. Djerourou: None. **M. Ptito:** None. **M. Vanni:** None.

Poster

PSTR149. Functional Organization of the Developing and Mature Visual System

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR149.20/AA16

Topic: D.06. Vision

Support: Japan Society for the Promotion of Science (JSPS) KAKENHI (JP21H03789, H.T.)
NSF 1457291(E.E.H.)

Title: Inter-species comparison of the vertical occipital fasciculus across mammalian species: a diffusion MRI study

Authors: *H. TAKEMURA[1,2,3], T. KANEKO[1,4], C. C. SHERWOOD[5], G. A. JOHNSON[6,7], M. AXER[8], E. E. HECHT[9], F. Q. YE[10], D. A. LEOPOLD[10];
[1]Natl. Inst. for Physiological Sci., Okazaki, Japan; [2]The Grad. Inst. for Advanced Studies, SOKENDAI, Hayama, Japan; [3]3.Center for Information and Neural Networks (CiNet), Advanced ICT Inst., Natl. Inst. of Information and Communications Technol., Suita, Japan; [4]Ctr. for the Evolutionary Origins of Human Behavior, Kyoto Univ., Inuyama, Japan; [5]Dept. of Anthrop., George Washington Univ., Washington, DC; [6]Duke Med. Ctr., Durham, NC; [7]Dept. of Biomed. Engin., Duke Univ., Durham, NC; [8]Res. Ctr. Jülich, Jülich, Germany; [9]Dept. of Human Evolutionary Biol., Harvard Univ., Cambridge, MA; [10]Natl. Inst. of Mental Hlth., Bethesda, MD

Abstract: The primate visual system is composed of dorsal and ventral cortical streams, which are specialized to serve spatial and object processing, respectively. While to some extent distinct, the two cortical streams are interconnected through prominent fiber tracts. For instance, the vertical occipital fasciculus (VOF) is a prominent white matter tract bridging dorsal and ventral visual areas lying superficial to the optic radiation (OR). The VOF has been demonstrated in humans and other primates through both classical neuroanatomical and modern neuroimaging studies (Wernicke, 1881; Yeatman et al., 2014; Takemura et al., 2017). To understand the nature and evolution of the VOF, we used a comparative approach, analyzing high-resolution, ex-vivo diffusion MRI (dMRI) data acquired from the brains of select mammalian species, including anthropoid and strepsirrhine primates, the tree shrew, and several species of rodents and carnivores. Most dMRI data were acquired by using 7T Bruker MRI in the National Institutes of Health, except for the rat and fox dMRI data provided by Duke University and Harvard University, respectively, and already analyzed in previous works (Johnson et al., 2021; Hecht et al., 2021). In each species, we first identified the OR using tractography and subsequently investigated diffusion signals in adjacent white matter. In all primate species tested (two cercopithecid monkeys, two platyrrhine monkeys, and two strepsirrhines), the OR was flanked laterally by a white matter compartment with a superior-inferior orientation perpendicular to the OR, demonstrating the existence of the VOF in each species. In contrast, there was no evidence of a white matter tract lateral to the OR in dMRI data acquired from the brains of four non-primate species in the Euarchontoglires superorder (tree shrew, ground squirrel, paca, and rat), nor high-resolution polarized light imaging data of the rat brain provided by Research Centre Jülich (Schubert et al., 2016; Zilles et al., 2016). In two carnivore species (ferret and fox), a small fiber compartment lateral to OR was tentatively identified but was much less prominent than VOF in primates. Taken together, our results demonstrate that the VOF is a conserved pathway among primate species but less prominent or absent in non-primate species. The expansion of the VOF may support primates' large behavioral repertoire involving visually directed actions toward objects, including manual reaching and grasping, physical interactions with conspecifics, and locomotion through an arboreal environment.

Disclosures: H. Takemura: None. **T. Kaneko:** None. **C.C. Sherwood:** None. **G.A. Johnson:** None. **M. Axer:** None. **E.E. Hecht:** None. **F.Q. Ye:** None. **D.A. Leopold:** None.

Poster

PSTR150. The Organization and Function of Visual Pathways To and From the Cortex

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR150.01/AA17

Topic: D.06. Vision

Support: NIH/NEI U01EY025858-04
NIH/NEI R01EY031589-01

Title: Quantifying retinal functional health in patients with varying scotoma patterns by accounting for cortical magnification factor

Authors: *E. CUTTS[1], M. MANIGLIA[2], P. DEMIRAYAK[1], M. DEFENDERFER[1], K. VISSCHER[1], D. DECARLO[1];
[1]Univ. of Alabama, Birmingham, Birmingham, AL; [2]Univ. of California Riverside, Riverside, CA

Abstract: Currently the leading cause of vision loss in older adults, macular degeneration is an eye disease that leads to death of photoreceptors in localized portions of the retina. This results in variable scotoma patterns from patient to patient. Complex visual tasks such as reading or navigation require both visual input as well as high-level visual processing. Patients differ wildly in their performance on complex visual tasks, even in patients with similar retinal damage. This suggests that high level visual processing differs between participants, but this difference has been challenging to quantify. As a step toward accurate quantification of high level processing, we developed a metric describing functional retinal health. It is not appropriate to define retinal functional health using typical performance measures such as acuity or contrast sensitivity because participants with extensive scotomas just outside the fovea can have excellent scores. On the other hand, methods like Macular Integrity Assessment (MAIA) give a more comprehensive view, as they examine perceptual thresholds at many locations across the retina. However, it has been unclear how to quantify MAIA images in order to compare visual performance between patients, whose patterns of scotoma can vary widely. We sought a measure of retinal functional health which appropriately weights spared central vision more strongly than spared peripheral visual locations. Cortical areas V1, V2, and V3 are retinotopically mapped, meaning that different portions of cortex correspond to different portions of vision. Less cortical area is devoted to portions of the retina responsible for peripheral vision than central vision; this means that central vision has a higher cortical magnification factor than does peripheral vision. By weighting the scores obtained on the MAIA by the cortical magnification factor of that part of vision, we account for the relative ‘importance’ of different portions of the visual field for performing a range of visual tasks. This retinal functional health measure predicts how an individual’s scotoma pattern influences performance on visual tasks. Here we describe validation of the technique, and argue that our measure gives a reliable value of retina functional health that can be used to better understand individual differences in performance for participants with low vision. This framework allows us to estimate the components of a participant’s performance on

complex visual tasks that can be explained by retinal functional health. In turn, this allows estimation of the degree to which a participant's performance relies on high-level visual processing.

Disclosures: **E. Cutts:** None. **M. Maniglia:** None. **P. Demirayak:** None. **M. Defenderfer:** None. **K. Visscher:** None. **D. DeCarlo:** None.

Poster

PSTR150. The Organization and Function of Visual Pathways To and From the Cortex

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR150.02/AA18

Topic: D.06. Vision

Support: AFOSR FA9550-21-1-0230
NSF 1718991

Title: A model of cortical error-correction from noisy retinal ganglion cell output

Authors: *A. BELSTEN[1,2], B. A. OLSHAUSEN[1,3,2];
[1]Herbert Wertheim Sch. of Optometry & Vision Sci., [2]Redwood Ctr. for Theoretical Neurosci., [3]Helen Wills Neurosci. Inst., Univ. of California, Berkeley, Berkeley, CA

Abstract: Efficient coding theory hypothesizes that sensory systems are adapted to input statistics so as to encode data in a way that maximizes its transfer to downstream areas while minimizing energy consumption and other neural resources. Karklin & Simoncelli [1] introduced a model based on this theory to account for functional properties of retinal ganglion cells (RGCs) in terms of joint source and noisy-channel coding, which has since been extended to explain other aspects of retinal coding [2]. However, the model lacks an explicit decoding mechanism to retrieve the original image data from its neuronal representation. In this study, we evaluate the efficacy of different decoding methods, including maximum a posteriori (MAP) estimation, linear decoding, and a fully unsupervised sparse coding model, in terms of their ability to reconstruct the original stimulus.
We show that both MAP estimation and linear decoding obtain a similar level of good signal reconstruction, relative to the RGC noise level. However, the computations involved in MAP estimation are complicated and biologically implausible. While linear decoding is simpler, it relies on access to the original images to learn the decoding weights. Sparse coding [3] has been proposed as a model of inferential computation in visual cortex that is adapted to higher-order statistical regularities. The model uses its learned representation to infer image structure from noisy input. Here we show that when trained on the simulated noisy outputs of an optimized model of retinal ganglion cell coding, sparse coding learns the patterns contained in these signals. Using these patterns as a prior over image structure, it can achieve reconstructions as good or better than both MAP estimation and linear decoding. By applying spike-triggered averaging to the sparse coding model's V1 cells, we recover the typical localized, oriented, and

bandpass receptive fields shown in previous work, but here obtained purely from training on simulated noisy on- and off-RGC activity.
Taken together, the results of this study provide a novel integration of theories of retinal and cortical function, demonstrating that the functional properties of a combined retina-V1 system can be accounted for in terms of efficient coding and Bayesian inference principles. Establishing this link is an important step in advancing our understanding of how information is processed and transformed across different stages of visual processing.
[1] Karklin & Simoncelli, Advances in neural information processing systems, 2011 [2] Roy, et al., Nature, 2021 [3] Olshausen & Field, Vision research, 1997

Disclosures: **A. Belsten:** None. **B.A. Olshausen:** None.

Poster

PSTR150. The Organization and Function of Visual Pathways To and From the Cortex

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR150.03/AA19

Topic: D.06. Vision

Support: Deutsche Forschungsgemeinschaft (SFB 870)
Max-Planck-School of Cognition
Hertie-Stiftung

Title: Deciphering thalamo-cortical activation at single-synapse resolution

Authors: *Y. CHEN[1], M. KLOOS[1], Y. ZHANG[1], I. PIRO[1], Z. VARGA[1], J. RICHTER[1], I. NELKEN[2,3], A. KONNERTH[1];
[1]Inst. of Neuroscience, TUM, Munich, Germany; [2]The Edmond and Lily Safra Ctr. for Brain Sci., [3]Dept. of Neurobio., The Hebrew Univ. of Jerusalem, Jerusalem, Israel

Abstract: Classical work by Hubel and Wiesel has established that in several mammalian species, orientation selectivity in the primary visual cortex (V1) is determined by the specific arrangement of non-tuned thalamic lateral geniculate nucleus (LGN) inputs to the cortical entry stage in layer 4 (L4). While evidence from different laboratories supports a similar model also for mouse V1, several other studies indicated the presence of orientation- and direction-tuned LGN neurons, at least in mice. Thus, the synaptic and circuit mechanisms contributing to orientation selectivity in mouse V1 may be more complex than initially assumed. In the present study, we established methods for in vivo imaging of single synapses that receive LGN inputs in L4 of mouse V1. To sparsely label orientation-tuned V1 neurons, we performed a two-step experiment in which population imaging with a synthetic calcium indicator (Cal-520) was followed by the delivery of plasmid vectors of genetically encoded calcium indicators (GECI) to orientation-tuned neurons via targeted single-cell electroporation. This experimental protocol allowed us to chronically monitor calcium signaling in spiny dendrites of such L4 neurons over several days. To distinguish thalamo-cortical (TC) synapses from cortico-cortical (CC) synapses,

we silenced V1 by optogenetic activation of Parvalbumin (PV) interneurons. Surprisingly, calcium imaging revealed mostly highly tuned CC synapses but provided no evidence for active TC single spine inputs. Therefore, we turned to glutamate imaging using the recently described iGluSnFR3 (Aggarwal et al, 2023). By combining glutamate imaging and cortical silencing, we were able to identify TC synapses in L4 neurons. We found that, in general, individual TC synapses were highly reliable, with large glutamate responses for every direction of the drifting-grating stimulus. In contrast, CC inputs were less reliable and highly tuned. Overall, the most striking new features of TC synapses in L4 neurons include (1) absence of postsynaptic calcium signaling, perhaps indicating the minor role of NMDA receptors, (2) the virtual absence of contributions from strongly orientation-tuned LGN neurons and (3) the presence of spatial clusters of synchronous TC single-spine inputs that may originate from the same afferent axon. In conclusion, we identified fundamental properties of TC synapses to L4 neurons, each of which representing upstream LGN neurons with characteristic receptive fields. Our results validate some of the basic aspects of Hubel and Wiesel theory on single-input level and add information on specific mechanisms of TC synapses.

Disclosures: **Y. Chen:** None. **M. Kloos:** None. **Y. Zhang:** None. **I. Piro:** None. **Z. Varga:** None. **J. Richter:** None. **I. Nelken:** None. **A. Konnerth:** None.

Poster

PSTR150. The Organization and Function of Visual Pathways To and From the Cortex

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR150.04/AA20

Topic: D.06. Vision

Support: NIH Grant EY030291
NIH Grant EY028905

Title: Directionally selective LGN neurons provide synaptic input to visual cortical simple cells with a similar direction preference

Authors: R. F. PLATT[1], C. SU[1], J.-M. ALONSO[2], H. A. SWADLOW[1], ***Y. BERESHPOLOVA**[1];
[1]Dept. of Psychological Sci., Univ. of Connecticut, Storrs, CT; [2]Biol. and Vision Sci., SUNY Col. of Optometry, New York, NY

Abstract: The rabbit retina contains ganglion cells that exhibit a high degree of selectivity for the direction of visual motion. These directionally selective retinal ganglion cells convey directional signals through the lateral geniculate nucleus (LGN) of the thalamus to the primary visual cortex (V1). Approximately 5-10% of neurons in the rabbit LGN show a strong preference for the direction of visual stimuli. We have previously shown that these LGN directionally selective (DS) cells generate a strong and focal synaptic drive in primary visual cortex (V1) that is similar in its laminar profile to that generated by LGN concentric cells, and that LGN DS cells

activate putative fast-spike inhibitory interneurons in layer 4 (L4). Notably, these cells lack directional selectivity, and also receive profuse input from LGN concentric cells. By contrast, the great majority of simple cells in layer 4 are highly selective to both stimulus orientation and direction, but how they achieve their directional preference and whether LGN DS neurons contribute to this preference, have been subjects of debate. Here we show that LGN DS neurons do contribute to the directional selectivity of L4 simple cells. In awake rabbits, we used cross-correlation methods to examine connectivity between LGN DS neurons and L4 simple cells of V1 and establish rules that govern this connectivity. We show that there is a high specificity in the connections between the simple cells and their thalamic inputs. Firstly, all connected thalamocortical pairs were highly retinotopically aligned, with distance between thalamic and cortical receptive field (RF) centers less than 1/2 the diameter of the LGN RF. Secondly, the amplitude of the postsynaptic local field potential generated by the LGN DS neuron near the simple cell is another necessary condition for ensuring synaptic connectivity between LGN DS/simple cell pairs. And lastly, LGN DS neurons form functional synaptic connections with simple cells only when their directional preferences are closely aligned. Thus, 7 of 10 cases were synaptically connected when LGN and cortical simple cell were retinotopically aligned (< 1/2 LGN RF separation) and directionally aligned within 30 degrees. By contrast none of 13 retinotopically aligned pairs were synaptically connected when direction alignment was off by > 60 degrees ($p<0.001$, Fisher's exact test). Our results demonstrate that LGN DS neurons provide directional information to directionally aligned V1 simple cells and do not provide directional information to simple cells that are not directionally aligned.

Disclosures: **R.F. Platt:** None. **C. Su:** None. **J. Alonso:** None. **H.A. Swadlow:** None. **Y. Bereshpolova:** None.

Poster

PSTR150. The Organization and Function of Visual Pathways To and From the Cortex

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR150.05/AA21

Topic: D.06. Vision

Support: NIH Grant MH099045
NIH Grant MH121841
NIH Grant EY022951
NIH Grant MH113852
Simons Foundation SFARI Research Grant
SMS Research Foundation
LouLou Foundation
NIH Grant DP1-EY033975
CDKL5 Forum Junior Fellowship Award

Title: Impact of CDKL5 deficiency on state-dependent thalamocortical dynamics

Authors: *R. L. OREN[1], M. J. HIGLEY[2], J. A. CARDIN[2];
[1]Interdepartmental Neurosci. Program, [2]Dept. of Neurosci., Yale Univ., New Haven, CT

Abstract: CDKL5 Deficiency Disorder (CDD) is an X-linked genetic disorder caused by mutations in the *cdkl5* (cyclin-dependent kinase-like 5) gene. Patients with CDD display features common to Autism Spectrum Disorder, but also frequently exhibit central visual impairments, including deficits in visually-evoked potentials, an indirect measure of thalamocortical input. Moreover, loss of CDKL5 is associated with epilepsy, a marker of cortical network hyperexcitability. Together, these phenotypes suggest that developmental loss of CDKL5 may shift the balance between thalamocortical and intracortical processing. Mice lacking CDKL5 recapitulate many human CDD phenotypes, including visual impairment, and may also exhibit dysregulation of arousal states. Work from our lab and others has demonstrated that arousal states reflect distinct physiological processes that influence spontaneous and sensory-evoked neural activity. However, the relative contribution of arousal state dysregulation to altered thalamocortical dynamics in models of CDD remains unclear. Furthermore, the impact of a shift from feedforward to recurrent intracortical network connectivity on visual processing is unknown. We recently developed techniques for dual-color mesoscopic imaging that enable us to simultaneously monitor spontaneous and visually-evoked thalamic axon activity in the primary visual cortex (V1) and cortical activity across the dorsal neocortex in the awake, behaving mouse. Using this approach, we have begun to characterize the functional consequences of CDKL5 deficiency for state-dependent thalamocortical dynamics and visual processing. Our preliminary data suggest that CDKL5 deficiency impacts arousal states in a sex-dependent manner, with mutant males exhibiting a hyperarousal phenotype and mutant females trending towards a hypo-arousal phenotype. However, both male and female CDKL5 deficient animals display increased cortical activity, particularly in frontal regions, and increased intracortical functional connectivity compared to littermate controls. We further find that CDKL5 deficient mice exhibit reduced visual response magnitudes in V1. Lastly, we find that spontaneous and visually evoked activity of thalamocortical axons are state-dependent and that correlations between thalamic activity and cortical visual area activity increase during periods of high arousal. Together, our results introduce a new method for interrogating thalamocortical inputs to V1 and provide unprecedented insight into the functional consequences of CDKL5 deficiency for state-dependent thalamocortical networks and visual processing in a mouse model of CDD.

Disclosures: R.L. Oren: None. **M.J. Higley:** None. **J.A. Cardin:** None.

Poster

PSTR150. The Organization and Function of Visual Pathways To and From the Cortex

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR150.06/AA22

Topic: D.06. Vision

Support: R01EY028905
R21EY030291

Title: Three types of Corticotectal neurons in rabbit visual cortex

Authors: *C. SU[1], R. F. PLATT[1], J.-M. ALONSO[2], H. A. SWADLOW[1], Y. I. BERESHPOLOVA[1];
[1]Univ. of Connecticut, Storrs, CT; [2]SUNY Col. of Optometry, New York, NY

Abstract: Impulses of corticotectal (CTect) axons generate a fast-conducting and powerful synaptic drive in the superficial layers of the superior colliculus (SC) (Bereshpolova et al., 2006). However, the functions of CTect pathways remain largely unknown, in part because of our limited understanding of the information conveyed by CTect neurons to the SC. Here we studied V1 CTect neurons in awake rabbits identified antidromically following electrical stimulation of the SC. We used sparse noise and drifting gratings to measure their spatial and temporal receptive fields (RFs) and examined the relationship between CTect RFs, the linearity of spatial summation and axonal conduction velocities. We demonstrate that, in rabbits, the RFs of *all* CTect neurons show a pronounced nonlinear spatial summation (F1/F0 <1) that makes the entire pathway poorly selective to the phase of grating patterns. In some classification schemes, this property alone would be sufficient to classify all CTect neurons as "complex" cells (Skottun et al., 1991). However, the spatial RFs structures of CTect neurons are heterogeneous in their spatial integration of inputs from ON and OFF visual channels and form three distinct types: (1) those with highly overlapping ON and OFF subfields (similar to classic "complex" cells (Hubel and Wiesel, 1962)), (2) those with highly separated ON and OFF subfields (similar to classic "simple" cells), and (3) those with a single ON or OFF subfield. Among the three types, type 1 sends the fastest conducting descending inputs to SC, type 2 has the sharpest orientation and direction selectivity, and type 3 contains the slowest conducting descending axons to SC. We also identified, using cross-correlation methods, a subpopulation of CTect neurons that receive potent monosynaptic input from single retinotopically aligned LGN neurons. These CTect neurons had a distinct depth profile and were among those with the fastest cortico-collicular axonal conduction times. We conclude that corticotectal pathways are very homogeneous in their non-linear responses to drifting visual gratings, but very diverse in their spatial responses to dark and light features in visual scenes, and in their synaptic connectivity with the LGN.

Disclosures: C. Su: None. **R.F. Platt:** None. **J. Alonso:** None. **H.A. Swadlow:** None. **Y.I. Bereshpolova:** None.

Poster

PSTR150. The Organization and Function of Visual Pathways To and From the Cortex

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR150.07/AA23

Topic: D.06. Vision

Title: Comparison of the visual discharge properties of primate superior colliculus and primary visual cortex neurons

Authors: *Y. YU, A. BOGADHI, Z. HAFED;
Univ. of Tuebingen, Tuebingen, Germany

Abstract: The primary visual cortex (V1) and superior colliculus (SC) both play important roles in the visual processing hierarchy. These areas are also anatomically and functionally connected, with the SC receiving direct inputs from V1. However, a detailed neurophysiological comparison of the visual neuronal response properties between these two brain areas remains untouched, with the general assumption being that SC neurons simply inherit the tuning properties of V1 neurons. Here, we recorded neuronal responses from the SC and V1 of two male rhesus macaque monkeys. The animals were trained to fixate, and we presented gabor gratings of various spatial frequencies and orientations for 300 ms over a gray background. The gratings were sized to approximately fill the SC visual response field (RF). To map the RFs, we first performed an initial RF mapping task using small white spots appearing over a gray background during fixation. We recorded from neurons preferring ~5-10 deg eccentricity, and we targeted lower visual field representations in the two areas. We also often recorded the two areas simultaneously, with overlapping RF locations. We found that SC neurons had significantly larger RF sizes than V1 neurons. The baseline activities of SC neurons were also, in general, higher than those in V1 neurons. V1 neurons, on the other hand, responded to stimulus onsets consistently earlier than simultaneously recorded SC neurons, and V1 neurons exhibited offset responses (at stimulus removal at the end of a trial) more robustly than SC neurons. As for feature tuning properties, SC orientation tuning widths were significantly larger than in V1, indicating weaker orientation selectivity. Spatial frequency tuning widths were more similar, but SC neurons preferred lower spatial frequencies in general. Interestingly, the first spike latencies of SC neurons increased systematically with image spatial frequency, independent of the neuron's preferred spatial frequency in terms of visual response strength (consistent with our earlier observations; Chen et al., 2018). However, despite responding earlier than SC neurons, V1 neurons did not show this temporal relationship with spatial frequency, nor did they show the dissociation with response sensitivity. These results demonstrate that the SC does not merely inherit V1 response properties, but that it is functionally specialized, perhaps to best exploit its proximity to the motor periphery. The SC also actively delays its responses to some stimuli despite the connected V1 having already spiked to these very same images.

Disclosures: Y. Yu: None. **A. Bogadhi:** None. **Z. Hafed:** None.

Poster

PSTR150. The Organization and Function of Visual Pathways To and From the Cortex

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR150.08/AA24

Topic: D.06. Vision

Support: NIH Grant NS118960
NIH Grant NS100016
NSF EPSCOR Award #1632738

Title: Parallel corticothalamic pathways from postrhinal cortex to pulvinar: synaptic and physiological properties

Authors: *J. B. ZALTSMAN[1], R. D. BURWELL[2], B. W. CONNORS[1];
[1]Neurosci., [2]Cognitive Linguistic & Psychological Sci., Brown Univ., Providence, RI

Abstract: Nearly all sensory information passes through the thalamus en route to the cortex. The neocortex in turn provides robust projections to the thalamus, with descending corticothalamic (CT) axons outnumbering thalamocortical (TC) axons by about 10:1. With these CT pathways, cortex likely exerts a considerable influence on the thalamus, and thus on its own inputs. Studies of CT pathways exploring inputs from unimodal primary sensory areas and the prefrontal cortex have revealed general motifs of anatomy and physiology, raising the question: Does every cortical area have a similar pattern of feedback to the thalamus? The parahippocampal cortex (postrhinal cortex, POR, in rodents), is a polymodal association area and the principal source of visual information to the hippocampus. The POR is heavily interconnected with the pulvinar nucleus of the thalamus (also known as the lateral posterior nucleus in rodents). Here, we utilize cortical layer-specific Cre- mouse lines, optogenetics and whole-cell recordings in a slice preparation as well as retrograde tracers and histology to investigate the physiological properties and anatomy of the top-down projections from cortical area POR to the thalamic pulvinar nucleus. We also use whole-cell recordings from across pulvinar neurons to explore the heterogeneity of intrinsic membrane properties. Consistent with the projection layers of other CT systems studied thus far, we observe retrogradely labeled neurons in layers 5 and 6 of POR. These retrogradely labeled cells overlap with cells expressing reporters in layer-specific Cre-mouse lines (Rbp4-Cre and Ntsr1-Cre). Using these lines, we investigated the synaptic and intrinsic properties of CT projections by recording cells in the pulvinar while optogenetically stimulating the terminals of CT cells. We find that Rbp4-Cre-targeted layer 5 cells evoke weakly depressing synaptic responses in pulvinar cells, while Ntsr1-Cre-targeted layer 6 cells evoke facilitating responses, again consistent with previous studies of primary sensory CT pathways. Our work suggests that the general structure and physiology of CT circuits are widely preserved across functionally diverse CT systems.

Disclosures: J.B. Zaltsman: None. **R.D. Burwell:** None. **B.W. Connors:** None.

Poster

PSTR150. The Organization and Function of Visual Pathways To and From the Cortex

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR150.09/AA25

Topic: D.06. Vision

Support: NIH R01 EY025219

Title: Functional ultrasound imaging reveals corticogeniculate feedback footprint in ferret lateral geniculate nucleus

Authors: *W. HU[1], S. ZHU[2], M. DOYLEY[1], F. BRIGGS[1];
[2]Univ. of Rochester, [1]Univ. of Rochester, Rochester, NY

Abstract: In the early visual system of highly visual mammals, visual information initially encoded in the retina, is further relayed to the visual thalamus (dorsal lateral geniculate nucleus or LGN) and then to primary visual cortex (V1). These feedforward retino-geniculo-cortical pathways have been widely studied using neurophysiological and neuroimaging methods. Complementing these feedforward pathways are corticogeniculate pathways that carry signals from V1 to LGN in the feedback direction. The function of corticogeniculate feedback in visual perception is less well understood. For example, the population-level influence of corticogeniculate feedback on LGN neurons, or the "footprint" of corticogeniculate influence among LGN neuronal populations, is not known. To address this question, we use optogenetics to manipulate the activity of corticogeniculate neurons in V1 combined with functional ultrasound imaging (fUS) to capture the footprint of corticogeniculate influence in the LGN. Experiments were performed in ferrets, highly visual carnivores with early visual system structure similar to that of primates. First, we inject genetically modified rabies virus (SADΔG-ChR2-mCherry) into LGN during surgery. Ten days after surgery, fUS is performed in anesthetized animals through a craniotomy over V1 and LGN using a Vantage 256 scanner (Verasonics, Inc.) with an 18 MHz L22-14vX transducer. Imaging is performed while animals view visual stimuli and an LED emitting 464nm light positioned over V1 to activate ChR2 expressed in infected corticogeniculate neurons is illuminated during some visual stimulation trials. Following imaging sessions, brain tissue is harvested for histological processing, including staining virus-infected corticogeniculate neurons. This enables 3D registration of the fUS imaged volume with the histological volume, in which infected corticogeniculate neuronal locations are marked, and an MRI ferret brain atlas (Hutchinson et, al 2017). Clusters of infected corticogeniculate neurons are used to identify regions of interest (ROIs) in fUS volumes. When comparing fUS signals in these ROIs across visual conditions with and without LED activation of corticogeniculate feedback, we find significant increases in fUS responses with LED activation of virus-infected corticogeniculate neurons within the same ROIs across multiple imaging planes and across multiple trials. These results demonstrate that fUS can capture optogenetic activation of CG neurons in V1. Additional experiments comparing retinotopic fUS activations in LGN with LED-activated corticogeniculate footprints in LGN are ongoing.

Disclosures: W. Hu: None. **S. Zhu:** None. **M. Doyley:** None. **F. Briggs:** None.

Poster

PSTR150. The Organization and Function of Visual Pathways To and From the Cortex

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR150.10/AA26

Topic: D.06. Vision

Support: Simons 542999

Title: Effects of locomotion on communication between visual areas in the mouse

Authors: *E. KIM[1], A. KOHN[4,2,3];
[1]Neurosci., [2]Opthalmology, [3]Systems and Computat. Biol., Albert Einstein Col. of Med., Bronx, NY; [4]Albert Einstein Col. of Med. Dominick P. Purpura Dept. of Neurosci., Albert Einstein Col. of Med. Dominick P. Purpura Dept. of Neurosci., Bronx, NY

Abstract: A central question in systems neuroscience is how brain states modulate sensory processing. Because sensory processing is accomplished by a distributed network of areas, how brain states modulate sensory processing might depend heavily on how they influence dynamic and flexible relaying of sensory signals between brain areas. Locomotion has been shown to affect visual representations at different stages of processing. However, how locomotion affects the relaying of signals between areas is poorly understood. We presented mice with visual stimuli both during locomotion and while they were still, whilst recording simultaneously from retinotopically-aligned portions of the dLGN, V1 and LM using high density laminar probes. Consistent with prior work, we found that locomotion enhanced firing rates of many neurons and improved encoding of visual stimuli, in each brain area. Encoding improvements were not evident with fluctuations in arousal and face movements, also monitored during our recordings. Trial-to trial response 'noise' correlations between neurons in different areas were weaker during locomotion—both for thalamocortical and corticocortical pairings. To understand how locomotion influenced population-based inter-areal signaling, we used multivariate linear regression models to predict activity in one area using activity in another. These models performed equally well for responses recorded while animals were still or locomoting. We then tested whether inter-areal signaling occurs through a communication subspace—a low-rank mapping between areas that relays some population activity patterns but not others. Both thalamocortical and corticocortical signaling occurred through communication subspaces, of similar dimensionality during still and locomoting periods. However, the communication subspaces measured during locomotion captured more of the available stimulus information in each area, suggesting they were better positioned to transmit visual information. To test this further, we identified an orientation coding axis in each area separately. The trial-to-trial projections of population activity onto these axes were more correlated across areas during locomotion. Our results suggest that locomotion can reconfigure inter-areal signaling, in addition to its effects on within-area representations of visual information.

Disclosures: E. Kim: None. **A. Kohn:** None.

Poster

PSTR150. The Organization and Function of Visual Pathways To and From the Cortex

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR150.11/AA27

Topic: D.06. Vision

Support: Shanghai Municipal Science and Technology Major Project (2018SHZDZX05)
he Strategic Priority Research Program of the Chinese Academy of Sciences(XDB32060000)

Title: Research about optogenetics manipulation on non-human primates

Authors: *D. LU;
Ctr. for Excellence in Brain Sci. and Intelligence Technol. , Chinese Acad. of Sci., Shanghai, China

Abstract: With the rapid progress of optogenetics, various genetic-modified mice models have been made to prosper the study in brain function. Given the fact that mice and human beings are quite different in anatomy, physiology, cognition, and behaviors, which means that to a certain extent, the conclusions derived from mice experiment may not be fully applied to explain some issues in human brain. Recently, there is a significant scarcity of monkey models that can be used for neuroscience. Here, we focused on genetically modified monkey models construction for neural manipulation and have obtained several kinds of transgenic monkey models. We used designed virus *pLVX-CamkII-ChRger2.0-EGFP* vector for high tilter lentivirus packaging to construct transgenic mice. Next, we performed histological validation on the obtained transgenic mice to ensure the expression of the selected opsin. In addition, after the histological analysis, we performed both intracellular and extracellular electrophysiological recording on the constructed transgenic mice to verify the function of the opsin. After the validation work in mice, we applied the same strategy to construct transgenic monkey models. We started with the transgenic mice construction. We injected 122 mouse embryos and totally got 42 survived individuals. Genotyping revealed a positive rate of 90.48%. Subsequently we applied the histological analysis in both genders, discovering that opsin was expressed across the whole brain in 34 survived individuals. After that we utilized intracellular recording to verifying the function of the opsin (n = 6), discovering that with continuous low light intensity stimulation, neurons with opsin expressing presented sensitive and stable channel opening (n = 30). To further assess the opsin functionality in vivo, we recorded the activity of pyramidal neurons from the anterior lateral motor cortex using 32-channel silicon probes (n = 5). During the recording, we detected that most neurons exhibited a significant increase in mean firing rate during the blue light stimulation (n = 60, $p < 0.05$, one-way ANOVA). Giving the promising results for neuronal activity manipulation in transgenic mice , we used the same strategy to generate transgenic monkey models. We totally obtained 9 survival individuals among which 6 were positive through genotyping. These results suggest that our strategy can be used for constructing transgenic models. Further experiments will be conducted to examine the practicability of our transgenic monkey models.

Disclosures: D. Lu: None.

Poster

PSTR150. The Organization and Function of Visual Pathways To and From the Cortex

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR150.12/AA28

Topic: D.06. Vision

Title: Nonsynaptic Muscarinic Receptor Localization on Extrastriate Feedback Axons in V1

Authors: *T. Q. NGUYEN, A. A. DISNEY;
Dept. of Neurobio., Duke Univ., Durham, NC

Abstract: Extrastriate feedback makes up a substantial portion of inputs to primary visual cortex (V1) and is hypothesized to contribute to surround suppression and top-down attention. Previous work has demonstrated that feedforward and local recurrent synapses are distinctly modulated by acetylcholine receptor subtypes, however it is not known how non-geniculate inputs are sculpted by acetylcholine.Here, we assessed whether muscarinic (m1 and m2) receptors are associated with V2 feedback axons or their synaptic partners in V1. Using immuno-electron microscopy and anterograde tracing of V2 projections to V1, we find that both types of muscarinic receptors are expressed on feedback axons from V2, but rarely, if ever at . Instead, these axons primarily exhibited non-synaptic or peri-synaptic expression of both muscarinic receptor types. Additionally, m1 and m2 receptors were found localized at cell bodies and presynaptic clefts of non-labeled cells, in agreement with previous anatomical work;. Nonsynaptic localization of muscarinic receptors has profound implications on how feedback axons, and the signals that they convey to V1, can be modulated by acetylcholine.

Disclosures: T.Q. Nguyen: None. **A.A. Disney:** None.

Poster

PSTR150. The Organization and Function of Visual Pathways To and From the Cortex

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR150.13/BB1

Topic: D.06. Vision

Support: DFG grant (ZA990/1-)
ZIAMH002838
ZIAMH002898
ZIAMH002899

Title: Direct comparison of electrophysiological- and fMRI-seed based functional connectivity in the macaque visual cortex

Authors: *D. ZALDIVAR[1], R. BHIK-GHANIE[2], D. C. GODLOVE[4], F. YE[3], D. A. LEOPOLD[5];
[1]Natl. Inst. of Mental Hlth., Bethesda, MD; [2]NIH, Natl. Inst. of Mental Hlth., Chevy Chase, MD; [3]Natl. Inst. of Mental Hlth., Besthesda, MD; [4]NIH, NIH, Bethesda, MD; [5]NIMH, NIMH, Bethesda, MD

Abstract: The correlational patterns of spontaneous fMRI activity across the brain are believed to reflect long-range network interactions among neural populations sharing intrinsic dynamics. Resting state functional connectivity (rs-fMRI) patterns overlap with known anatomical connectivity and has become a core method to infer the brain's organizational principles. However, as rs-fMRI is only an indirect measurement of neural activity, its bearing on the structure and physiology of neural networks is imprecise. For example, to what extent does rs-fMRI capture the large-scale functional interactions of individual neurons? To address this question, we concurrently measured whole-brain fMRI activity with local electrophysiological signals in five macaque monkeys. We recorded spiking and local field potential activity from electrodes implanted in different regions of the occipitotemporal pathway, including area V4, the anterior medial (AM) face patch, and the anterior fundus (AF) face patch. During periods of unstimulated rest, we compared rs-fMRI across the brain using electrophysiological versus fMRI seeds at each of the recording sites. While the two different seed strategies revealed some overlap in functional connectivity, they also revealed notable points of divergence. First, electrophysiological seed maps yielded more spatially restricted correlations than did fMRI seed maps. Second, fMRI seed maps were more bilaterally symmetrical. Third, electrophysiological seed maps included areas that were never revealed in fMRI seed maps, including thalamic and neuromodulatory structures. Fourth, electrophysiological seeds revealed demonstrated neurons with both positive and negative correlations with cortical areas for which the fMRI seed only showed positive correlations. These findings indicate that while fMRI functional connectivity captures some aspects of the brain's functional organization during rest, it (1) may overestimate the breadth and bilateral symmetry of cortical interactions and (2) is prone to miss other types of whole-brain circuit interactions that are evident through electrophysiological seed-based correlations. The direct linking of local electrophysiological signals to whole-brain fMRI across the brain offers new avenues to study how local neural signals contribute to the physiology of brain-wide neural circuits, during rest and behavior.

Disclosures: D. Zaldivar: None. **R. Bhik-Ghanie:** None. **D.C. Godlove:** None. **F. Ye:** None. **D.A. Leopold:** None.

Poster

PSTR150. The Organization and Function of Visual Pathways To and From the Cortex

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR150.14/BB2

Topic: D.06. Vision

Support: Europäischer Fonds für regionale Entwicklung
Deutsche Forschungsgemeinschaft
BBSRC(UK)
ESF
Studienstiftung des deutschen Volkes

Title: Histological validation of diffusion MRI and tractography for tracing fibre tracts in macaque extrastriate visual cortex.

Authors: *D. L. N. SAMS[1], J. E. T. SMITH[2], C. GAILLARD[1], A. BASHIR[3], H. BRIDGE[4], T. B. DYRBY[5], K. KRUG[1];
[1]Otto von Guericke Univ., Magdeburg, Germany; [2]Ernst Strüngmann Inst. (ESI) for Neurosci., Frankfurt, Germany; [3]Dept. of Physiology, Anat. and Genetics, Univ. of Oxford, Oxford OX1 3PT, UK., Oxford, United Kingdom; [4]Clin. Neurol., Univ. of Oxford, Oxford, United Kingdom; [5]Danish Res. Ctr. for Magnetic Resonance, Ctr. for Functional and Diagnos. Imaging and Research, Copenhagen Univ. Hospital, Amager & Hvidovre, Hvidovre, Copenhagen, Denmark

Abstract: Diffusion-weighted magnetic resonance imaging (dMRI) with probabilistic tractography can non-invasively map white matter connectivity in human and animal brains, but it remains unclear how well this method captures underlying fibre tracts. We quantitatively compare specific white matter tracts labelled from a small cortical tracer injection site with tractography streamlines generated from the same cortical site.Biotinylated dextran amine (BDA) tracer (200 nL) was placed into the left extrastriate visual area V5/MT of two anaesthetized macaque monkeys, one male (M127), one female (M131). Ten to fifteen days after the injection, monkeys (mean age 9.3 years) were perfused and *post mortem* dMRI were collected on a 4.7T Agilent pre-clinical MRI scanner using a 2D single spin-echo sequence with a single-line readout (voxel size 0.5 x 0.5 x 0.5 mm). Brains were sectioned parasagittally at 50 μm. Tracer paths were hand-drawn in Neurolucida in a 1-in-5 series of sections. Using each brain's fractional anisotropy (FA) images, probabilistic tractography was conducted using ProbtrackX2 from the FSL FDT toolbox seeding at the injection site. Histological data including section outline and grey-white matter boundaries were aligned with the dMRI FA image in MATLAB.Qualitatively, major streamline tracts aligned well with the labelled cortical projections from MST, LIP, and VIP, but not from V1 and V2 to V5/MT. Quantitatively, 2D spatial cross-correlation revealed considerable differences between the alignment of histological tracts and tractography streamlines in the anterior-posterior plane (mean drift: 1.9mm M127, 2.9mm M131) and dorsal-ventral plane (mean drift: 6.1mm M127, 7.0mm M131) in both brains.We applied Gaussian smoothing to tractography data and used receiver operating characteristic (ROC) curves to identify the overlap probability at different thresholds. We calculated the Area Under the Curve (AUC) and found reasonable overlap between tracer and tractography data (M127: AUC = 0.66, M131: AUC=0.80).Our results show that dMRI and tractography streamline can capture major aspects of specific white matter tract pathways, but major limitations still exist. The high curvature of tracts from V5/MT or towards V1 increases the likelihood of deviation from tracer data as distance from the seed point increases. Applying thresholds to tractography to include the top 99%, 95% or 90% of streamlines results in rapid data loss. Direct comparisons of high resolution dMRI data to underlying ground-truth histology can aid optimisation of data acquisition and tractography analysis to enhance the reliability and accuracy of non-invasive tracing of connections.

Disclosures: D.L.N. Sams: None. **J.E.T. Smith:** None. **C. Gaillard:** None. **A. Bashir:** None. **H. Bridge:** None. **T.B. Dyrby:** None. **K. Krug:** None.

Poster

PSTR151. Sensorimotor Transformations: Human and Nonhuman Primate

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR151.01/BB3

Topic: D.07. Visual Sensory-Motor Processing

Support: U.S. Department of Health & Human Services | NIH | National Eye Institute (NEI) - ZIAEY000570-01
Deutsche Forschungsgemeinschaft (German Research Foundation) - 276693517
National Science Foundation (NSF) - IIS1350990

Title: Modulation of activity in primate visual cortex by spontaneous movements is minimal during fixation and free-viewing

Authors: *B. TALLURI[1], I. KANG[1], A. LAZERE[1], K. R. QUINN[2], N. KALISS[1], J. YATES[3], D. A. BUTTS[4], H. NIENBORG[1];
[1]NIH, Natl. Eye Inst. (NEI), Bethesda, MD; [2]Ctr. for Integrative Neurosci., Univ. of Tuebingen, Tuebingen, Germany; [3]UC Berkeley, UC Berkeley, Oakland, CA; [4]Univ. of Maryland, Univ. of Maryalnd, College Park, MD

Abstract: Recent studies in mice reported brain-wide neuromodulation due to locomotion and spontaneous body movements. Surprisingly, this modulation by movements in early visual regions of the mouse was found to be larger than that due to the visual information from the retina. These results suggest an interaction between movements and early sensory processing.We set out to examine the extent to which sensory processing in the early visual cortex is susceptible to non-sensory variables in non-human primates, a species whose visual system closely matches that of humans. We recorded extracellularly from V1, V2 and V3/V3A (n=293, 351 and 312 units respectively) in two adult male rhesus monkeys performing visual tasks involving repeated presentations (trials) of visual stimuli. Monkeys were head-fixed and seated in primate chairs in an experimental setup directly comparable to one of the studies in mice, while their body and eye movements were monitored with multiple cameras. We used multivariate linear encoding models to quantify the unique contribution of task covariates, body movements, and slow fluctuations in neuronal activity to single-trial neural dynamics. The modulation of neural activity by spontaneous movements was absent when the retinal input was controlled: during stimulus epochs when animals fixated on a central spot and inter-trial intervals in which neuronal receptive fields (RFs) were within the uniformly-lit screen. For periods where RFs were outside the screen, adding eye-position and pupil regressors to the model largely removed the apparent contributions of spontaneous movements. We performed a follow-up experiment to test whether the absence of modulation by movements can be explained by the behavioral state of the animals during task performance. Here, the animal either fixated on a central spot (fixation blocks) or was freely-viewing (free viewing blocks), each block spanning 50-100 trials, as a sequence of full-screen luminance flashes were presented in quick succession. While neuronal responses showed clear modulation across blocks, the modulation of neural activity by spontaneous movements was minimal and indistinguishable between fixation and free-viewing blocks. Together, these results show that in contrast to the results in mice, in the primate visual cortex,

spontaneous movements minimally modulate neural activity irrespective of the behavioral state of the animal.

Disclosures: **B. Talluri:** None. **I. Kang:** None. **A. Lazere:** None. **K.R. Quinn:** None. **N. Kaliss:** None. **J. Yates:** None. **D.A. Butts:** None. **H. Nienborg:** None.

Poster

PSTR151. Sensorimotor Transformations: Human and Nonhuman Primate

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR151.02/BB4

Topic: D.07. Visual Sensory-Motor Processing

Support: H2020-EIC-FETPROACT-2019-951910-MAIA
Ministry of University and Research, Grant no. 20208RB4N9
Ministry of University and Research, NEXTGENERATIONEU
Ministry of University and Research, NATIONAL RECOVERY AND RESILIENCE PLAN (NRRP), PROJECT MNESYS (PE0000006)-DN.1553 11-10-2022

Title: Comparison of eye position activity in parietal cortex during goal directed and non-goal, self-generated eye movements: does the context matter?

Authors: *K. HADJIDIMITRAKIS, F. E. VACCARI, S. DIOMEDI, M. DE VITIS, M. FILIPPINI, P. FATTORI;
Univ. of Bologna, Bologna, Italy

Abstract: Neurons in posterior parietal cortex (PPC) control several effectors (e.g. eye, hand, foot) and encode space locations in a variety of spatial coordinate systems, including those centred relative to the eye position, and to the positions of the head, shoulder or hand. One of the characteristic properties of PPC is that its populations compute flexible yet stable representations of spatial locations and effector movements across various behavioural contexts that have been demonstrated in numerous studies involving goal-directed behaviors. However, little is known about the neural correlates of these representations during internally driven, non-goal-directed behaviors. We addressed this issue in medial PPC cortex (area V6A) one of the key nodes of the parietofrontal action network, which is involved in space coding and movement planning. We compared the eye position signals of single V6A neurons in macaque monkeys while they made saccades to fixate visual targets presented in various directions and distances in the peripersonal and extrapersonal space and with the gaze signals while they made spontaneous saccades in the dark before the visual targets appeared. We used methods to quantify the 3D eye position field and compare the spatial selectivity of gaze signals between the two behavioural contexts. We found that: a) the firing rates of V6A neurons were comparable in the two conditions, b) the spatial tuning of gaze activity was stronger after the spontaneous, no stimulus-driven, saccades compared to the visually-guided saccades. In sum, these data reveal unexpected differences in

the encoding of space during cue instructed and free movement behaviour in an area studied exclusively during visually-guided tasks. Accordingly, these findings highlight that several aspects of the parietofrontal network organization need to be revised.

Disclosures: K. Hadjidimitrakis: None. **F.E. Vaccari:** None. **S. Diomedi:** None. **M. De Vitis:** None. **M. Filippini:** None. **P. Fattori:** None.

Poster

PSTR151. Sensorimotor Transformations: Human and Nonhuman Primate

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR151.03/BB5

Topic: D.07. Visual Sensory-Motor Processing

Support: H2020-EIC-FETPROACT-2019-951910-MAIA
Ministry of University and Research (20208RB4N9)
NEXTGENERATIONEU (NGEU) and funded by the Ministry of University and RESEARCH (MUR), NATIONAL RECOVERY AND RESILIENCE PLAN (NRRP), PROJECT MNESYS (PE0000006)-DN.1553 11-10-2022)

Title: Exploring neural subspaces in Posterior Parietal Cortex during planning and reaching movements

Authors: *S. DIOMEDI, F. E. VACCARI, M. DE VITIS, M. FILIPPINI, P. FATTORI; Univ. of Bologna, Bologna, Italy

Abstract: The posterior parietal cortex (PPC) is widely acknowledged as a crucial brain region involved in the planning and execution of reaching movements. Our research has provided compelling evidence that challenges the ability to categorize distinct subpopulations of neurons based on their functions, revealing a phenomenon known as mixed selectivity (Diomedi et al., 2020). Additionally, a sequence of neural states has been found, that aligns with the different motor stages of a reaching task. Furthermore, we have demonstrated that each neural state relies on the collective activity of the entire neuronal population rather than being driven by specific subpopulations, effectively refuting the notion of a state-specific class of neurons (Diomedi et al., 2021). Thus, the same neurons perform different computations depending on the movement phase, raising questions about how the entire population organizes its activity to allow this flexibility.Here, we characterize the neural subspaces in three PPC areas, namely V6A, PEc, and PE (monkey1, [134 116 48]; monkey2 [99 84 82]), during the planning and movement phase of a reaching task. A preliminary analysis reveals a time-varying correlation structure within the neural populations, suggesting a series of non-overlapping subspaces in which the spiking activity unfolds (average R2 for both animals: V6A 0.07, PEc 0.04, PE 0.03). Employing a manifold-optimized version of Principal Component Analysis, we identified two orthogonal subspaces during planning and movement, each capturing on average 90% of the variance across

monkeys and areas. However, cross-projections (i.e. projecting the movement activity in the planning subspace and vice versa) account for a significant amount of the variance (10%). This indicates an incomplete orthogonality between neural dynamics suggesting the presence of exclusive neural dimensions for each of the time epochs and shared dimensions between epochs. Indeed, we found that the exclusive subspaces captured around 80% of normalized variance, across animals and areas. On average, shared subspaces explained 28% of plan and movement related neural activity.In conclusion, we show here the existence of both shared and exclusive populational-level subspaces across the three parietal areas explored that may allow PPC neural activity to carry out different epoch-specific computations using the same neural substrate.

Disclosures: S. Diomedi: None. **F.E. Vaccari:** None. **M. De Vitis:** None. **M. Filippini:** None. **P. Fattori:** None.

Poster

PSTR151. Sensorimotor Transformations: Human and Nonhuman Primate

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR151.04

Topic: D.07. Visual Sensory-Motor Processing

Title: Hand actions without hands: effector independent representations in the visuomotor system

Authors: Y. LIU[1], F. A. MARTINEZ ADDIEGO[2], N. ZUR[4], S. SEN[3], M. RIESENHUBER[5], J. C. CULHAM[7], ***E. STRIEM-AMIT**[6];
[1]Inst. of Neuroscience, Key Lab. of Primate Neurobio., Chinese Acad. of Sci., Shanghai, China; [2]Georgetown Univ., Cabin John, MD; [3]Georgetown Univ., Washington, DC; [4]Georgetown Univ. Med. Ctr., [5]Dept Neurosci, [6]Georgetown Univ. Med. Ctr., Washington, DC; [7]Western Univ., Western Univ., London, ON, Canada

Abstract: We perform most of our daily actions with our hands, and vast portions of the action system in the brain are dedicated to them. What does this system do in people born without hands who use their feet instead? I'll present a series of fMRI experiments that show that many hand-selective brain regions do not represent the motor control of the hands themselves but rather what actions are performed or perceived with them for actions such as grasping, reaching, and tool use. These higher-level representations in the parietal and frontal cortices allow action planning that is indifferent to the body part performing the action and allow us to understand actions we cannot perform ourselves. Last, such research can open new avenues to use high-level action representations for motor rehabilitation and prosthesis design.

Disclosures: Y. Liu: None. **F.A. Martinez Addiego:** None. **N. Zur:** None. **S. Sen:** None. **M. Riesenhuber:** None. **J.C. Culham:** None. **E. Striem-Amit:** None.

Poster

PSTR151. Sensorimotor Transformations: Human and Nonhuman Primate

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR151.05/BB6

Topic: D.07. Visual Sensory-Motor Processing

Support: NIH Grant R21 NS118055
National Science Foundation Graduate Research Fellowship Program (NSF GRFP)
Rackham Merit Fellowship from the University of Michigan

Title: State-dependent effects of parietal stimulation on cortical plasticity and action control

Authors: E. GOLDENKOFF[1], J. A. DELUISI[2], T. G. LEE[1], J. A. BRISSENDEN[1], S. F. TAYLOR[3], T. A. POLK[1], *M. VESIA[4];
[1]Univ. of Michigan, [2]Univ. of Michigan, Ann Arbor, MI; [3]Univ. of Michigan Dept. of Psychiatry, Univ. of Michigan Dept. of Psychiatry, Ann Arbor, MI; [4]Univ. of Michigan, Ann Arbor, Ann Arbor, MI

Abstract: Repetitive transcranial magnetic stimulation (rTMS) is a widely used tool to modulate brain circuits, brain plasticity, and behavior non-invasively. Although rTMS affects interconnected brain networks, its impact remains unclear. The effects of rTMS on the brain and behavior are influenced by neural activity during stimulation. Thus, the brain state during stimulation may play a role in the lasting effects of rTMS on distal brain responses and associated behaviors. However, the consequences of directly manipulating brain activity over multiple days on brain function and behavior still need to be better understood. Therefore, our study aimed to investigate whether controlling behavior during multi-day rTMS targeting the grasp control region of the posterior parietal cortex (PPC) could enhance functional specificity in neuromodulation within the parietal-frontal network responsible for action control. We used intermittent theta burst stimulation (iTBS) on the PPC to explore the relationship between stimulation and brain state. In our study, 48 healthy volunteers were randomly assigned to one of three rTMS intervention groups. They received 4 days of iTBS, followed by electrophysiological and behavioral assessments. Group 1 received iTBS to PPC while performing a grasping task, Group 2 received iTBS to PPC while resting, and Group 3 received iTBS to a parietal region outside the grasping network while performing a grasping task. We compared changes in motor cortical excitability and performance before and immediately after the fourth rTMS session. We hypothesized that applying parietal rTMS during a grasping task over multiple days would increase motor excitability and improve performance compared to using the same stimulation protocol during rest. Our results confirmed that undergoing numerous sessions of targeted rTMS on the parietal network increased excitability in the downstream motor cortex. Moreover, we observed that stimulation combined with a grasping task further increased motor cortical excitability. Importantly, manual dexterity performance improved only following brain-state-controlled PPC stimulation. In conclusion, our findings suggest that using a behavioral task to regulate brain activity during network-targeted brain stimulation can enhance the process of plasticity induction in the cortical circuit mechanisms responsible for mediating action processes.

Disclosures: **E. Goldenkoff:** None. **J.A. Deluisi:** None. **T.G. Lee:** None. **J.A. Brissenden:** None. **S.F. Taylor:** None. **T.A. Polk:** None. **M. Vesia:** None.

Poster

PSTR151. Sensorimotor Transformations: Human and Nonhuman Primate

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR151.06/BB7

Topic: D.07. Visual Sensory-Motor Processing

Support: Leibniz Association: Collaborative Excellence - Neuro-Optogenetics (WGL SAW-2014-DPZ-1)
DFG CRC-889 "Cellular mechanisms of sensory processing"

Title: Delaying of context-specific sensorimotor transformations by pathway-specific optogenetic inhibition of frontal-to-parietal projections in rhesus monkeys

Authors: *H. GUO[1], M. G. FORTUNA[1,3], J. HÜER[2,4,5], S. TREUE[2,6,7,8], A. GAIL[1,6,7,8]; [1]Sensorimotor Group, [2]Cognitive Neurosci. Lab., German Primate Ctr., Goettingen, Germany; [3]Allen Inst. for Brain Sci., Seattle, WA; [4]Ernst Strüngmann Inst. for Neurosci. in Cooperation with Max Planck Society, Frankfurt, Germany; [5]Lab. de Neurosciences Cognitive et Computationnelles, Ecole Normale Supérieure, INSERM, PSL Univ., Paris, France; [6]Fac. for Biol. and Psychology, Univ. of Goettingen, Goettingen, Germany; [7]Leibniz ScienceCampus Primate Cognition, Goettingen, Germany; [8]Bernstein Ctr. for Computat. Neurosci. Goettingen, Goettingen, Germany

Abstract: Context-sensitive visuomotor transformations engage the frontoparietal network in primate cerebral cortex. It has been hypothesized that the dorsal premotor cortex (PMd) and the parietal reach region (PRR) coordinate their activities via reciprocal connections to select motor goals. However, the causal influence of PMd activity on neural dynamics in PRR during context-sensitive action selection remains unclear. We investigated PMd impact on PRR motor-goal encoding, hypothesizing a stronger effect during non-standard mapping (anti-reach) with higher cognitive control, comparing to direct mapping (pro-reach). We transduced neurons in the PMd area of two monkeys with the inhibitory opsin eArchT3.0 delivered using an AAV5 vector driven by the CaMKIIα-promoter (AAV2/5-CaMKIIα-eArchT3.0-eYFP) and applied pathway-selective (PMd to PRR) optogenetic partial silencing of PMd axons projecting to PRR, while monkeys performed a memory-guided, center-out, anti-reach task. We applied laser stimulation during the presentation of visual cues instructing the spatial target and the pro- or anti-reach task rule. Simultaneously, we conducted single-unit microelectrode recordings in the laser-stimulated neuropil, either in PMd or in PRR, in which only axons arriving from PMd should be transfected. Laser stimulation in PMd locally silenced PMd neural activity. Laser stimulation in PRR locally induced modulation of neural responses in individual neurons, indicating the efficacy of inhibiting presynaptic activity from PMd. These effects persisted during the subsequent movement planning period. At population level, directional selectivity of PRR spiking activities

was delayed with laser stimulation. The effect of silencing PMd inputs on PRR activity was context-dependent, as the delay in directional response was induced only during anti-reach trials. Directional decoding in PRR revealed a temporary increase in the visual-cue representation in laser-stimulated anti-trials, which suggested a reduced suppression of the pro-target representation. When simulating the pro-/anti- reach task with a modular recurrent neural network, including inhibitory input from PMd to PRR, we found equivalent neural dynamics and perturbation effects as when inhibiting the PMd-to-PRR input in the optogenetic experiment. Our results support the hypothesis that dynamic reorganization in PRR, as it is selectively needed for spatial remapping in anti- but not pro-reach trials, depends on the functional and direct input from PMd. Our findings support the notion that rule-based, goal-directed reaching partly builds on frontal-to-parietal causal modulation.

Disclosures: H. Guo: None. **M.G. Fortuna:** None. **J. Hüer:** None. **S. Treue:** None. **A. Gail:** None.

Poster

PSTR151. Sensorimotor Transformations: Human and Nonhuman Primate

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR151.07/BB8

Topic: D.07. Visual Sensory-Motor Processing

Support: IBS-R015-D1
2023R1A2C2005290

Title: The impact of motion direction expectations on neural activity in the frontal eye field during smooth pursuit eye movements

Authors: *S. KIM[1,4], J. PARK[5,4], J. LEE[4,2,3];
[1]Sungkyunkwan Univ., Suwon, Korea, Republic of; [2]Sungkyunkwan Univ., Gyeonggi-Do, Korea, Republic of; [3]Dept. of Intelligent Precision Healthcare Convergence, Sungkyunkwan Univ., Suwon, Korea, Republic of; [4]Institue for Basic Sci. (IBS), Suwon, Korea, Republic of; [5]Neurosciences, Washington Univ. in St. Louis, St. Louis, MO

Abstract: Bayesian inference aids us in making decisions in uncertain situations by merging our existing beliefs with new evidence. The function of Bayesian inference in our brain is an ongoing topic of research, as our understanding of the exact mechanisms is not yet comprehensive. In our previous study, we investigated the effect of prior expectations on neural representation in the middle temporal area (area MT/V5) using smooth pursuit eye movements as a behavioral assay. Our findings demonstrated that prior expectations have a diminishing effect on neural responses in area MT, with the reduction depending on their preferred directions when the sensory evidence was weak. This decrease in response effectively honed the direction tuning of the neural population, leading to less variation in pursuit directions across trials. Building upon these findings, our current study shifts the focus from area MT to the frontal eye field (FEF) to

examine the effect of prior expectations on neural activity in a higher-order brain region. We recorded neural activities from 457 and 124 cells in the FEF of two rhesus monkeys while they performed a smooth pursuit eye movement task, where we manipulated both the strength of sensory motion and the prior knowledge of motion direction. Our results revealed that while prior expectations only reduced the neural response in area MT when sensory evidence was weak, they consistently reduced FEF neural responses regardless of the reliability of sensory evidence. Notable, this trend was also observed in spontaneous activity, with statistically significant results observed in one monkey. The findings from our earlier study in area MT suggest that this sensory area may represent the posterior distribution within the Bayesian inference framework. Our current findings further support the notion that the FEF may serve as a source of prior information, delivering task-specific expectation signals to area MT. These discoveries significantly contribute to our understanding of how prior expectations modulate neural activity in different brain regions within the hierarchical Bayesian inference framework during smooth pursuit eye movements.

Disclosures: S. Kim: None. **J. Park:** None. **J. Lee:** None.

Poster

PSTR151. Sensorimotor Transformations: Human and Nonhuman Primate

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR151.08/BB9

Topic: D.07. Visual Sensory-Motor Processing

Support: National Key R&D Program of China (Grant 2020YFB1313402)
National Key R&D Program of China (Grant 2017YFA0701102)
National Science Foundation of China (Grant 31871047)
National Science Foundation of China (Grant 31671075)
Shanghai Municipal Science and Technology Major Project (Grant 2021SHZDZX)

Title: Predictive coding of tracking and intercepting inferred moving target in posterior parietal cortex

Authors: *R. ZHENG[1,2], Y. GU[1], H. CUI[1,2];
[1]Ctr. for Excellence in Brain Sci. and Intelligence Technol. (Institute of Neuroscience), Shanghai, China; [2]Chinese Inst. for Brain Res., Beijing, China

Abstract: To generate appropriate action in the changeable environment, compensating for inevitable delays in sensory transmission and movement execution is essential, which makes prediction integral to the sensorimotor transformation. Accurate visual-guided hands movement is a distinctive characteristic of the primate, which is accompanied by the complication of the sensorimotor system, especially the posterior parietal cortex (PPC). Two subregions in PPC, the lateral intraparietal cortex (LIP) and the parietal reach region (PRR), have been identified as

areas involved in translating sensory information to saccades and reach action, respectively. To further elucidate neural mechanisms mediating estimation of current states of external stimuli and their consequences for an upcoming movement, single-unit activity was recorded from both LIP and PRR while monkeys were trained to perform an occluded manual interception task. In particular, during the cue period, a target first moved circularly on the visual display, for a random duration with one of five angular velocities. The target then disappeared for a fixed period of occlusion. Finally, a GO cue appeared and the monkey was required to intercept the target within a limited time window. Neural recordings revealed that as a trial proceeded, LIP and PRR exhibited a gradient from sensory-dominant tuning to movement-dominant. This transition took the form of a progressive sensory prediction combined with an abrupt motor prediction. Most neurons reflected the sensory signal in the cue period. Interestingly, some neurons not only encoded delayed or instantaneous target direction, but also extrapolated target direction into the near future. Once the target was masked, most cells were modulated by the forthcoming movement, although some still represented the inferred target direction. As the GO cue approached, the number of cells representing reach intention significantly increased, as well as the number of cells related to timing. Furthermore, the predictive type of tunings was likely to have larger tuning width, suggesting the dynamics of tuning properties may be a clue to reveal the predictive coding. During planning interception based on the inferred moving target, LIP preserved more actual and inferred sensory information, and PRR tended to form reach plan as early as possible. Our results suggest that the internal estimation of stimulus state and impending motor plan can be both embodied in PPC, revealing an essential role of PPC in motor control. On the other hand, the differences and commonalities between LIP and PRR suggest a functional separation and a continuum of predictive sensorimotor processing in PPC.

Disclosures: R. Zheng: None. **Y. Gu:** None. **H. Cui:** None.

Poster

PSTR151. Sensorimotor Transformations: Human and Nonhuman Primate

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR151.09/BB10

Topic: D.07. Visual Sensory-Motor Processing

Support: CIHR grant with neural recordings PJT-159559

Title: Partial convergence of proprioception and visual feedback across primary motor cortex

Authors: *M. T. JACOBS[1], K. P. CROSS[2], A. KUMAR[1], S. H. SCOTT[3,1];
[1]Ctr. for Neurosci. Studies, Queen's Univ., Kingston, ON, Canada; [2]Neurosci. Ctr., Univ. of North Carolina, Chapel Hill, NC; [3]Dept Biomed. and Mol. Sci., Queens Univ., Kingston, ON, Canada

Abstract: A key feature of the motor system is the ability to rapidly generate goal-directed motor corrections to counteract disturbances of the limb. Theoretical models of behaviour based

on optimal feedback control suggest different sources of sensory information (i.e., visual, proprioceptive) are integrated together before this limb-related information is passed to motor regions like motor cortex, which in turn, generate goal-directed motor corrections. Such models predict that the same neurons in motor cortex should respond to both proprioceptive and visual feedback when generating motor corrections. In contrast, the anatomical organization of sensorimotor cortex suggests there should be regional differences in sensory feedback to motor cortex. This suggests there should not be complete overlap of proprioceptive and visual feedback in motor cortex. Three male monkeys (Macaca mulatta) were trained to perform reaching movements using the Kinarm robotic lab. Finger position was represented with a cursor (circle) presented in a virtual reality system aligned with the horizontal workspace. On random trials, either the cursor was shifted (visual perturbation) or a mechanical load was applied to the limb (proprioceptive perturbation) requiring the monkey to make a motor correction to attain the spatial goal. Perturbations occurred perpendicular to the lateral reach movement (i.e., above, or below). In two monkeys, a 96-channel grid was placed on the of the arm region of motor cortex. In the third monkey, a chamber was implanted to allow individual micro-electrodes to be inserted transdurally permitting neural recordings across cortex including the bank of the central sulcus. The recordings from the arrays (n =257) identified that most neurons (~67%) responded to sensory feedback. Of responsive neurons (n=173), approximately half were multimodal (~54%), with the other half responding only to proprioceptive (~27%) or visual feedback (~19%). The response range between visual and proprioceptive perturbations were correlated (range of r= [0.68, 0.86]) and the neural subspaces containing the responses to each perturbation type overlapped (range of overlap index=0.36-0.47). Preliminary findings from micro-electrode recordings in the caudal bank of motor cortex suggest a similar distribution as found with the arrays located on the surface of motor cortex. The results display some amount of convergence between sensory modalities in motor cortex. However, there was a clear presence of many neurons that responded to only one sensory modality highlighting incomplete integration across visual and proprioceptive modalities at the level of motor cortex.

Disclosures: **M.T. Jacobs:** None. **K.P. Cross:** None. **A. Kumar:** None. **S.H. Scott:** E. Ownership Interest (stock, stock options, royalty, receipt of intellectual property rights/patent holder, excluding diversified mutual funds); S.H.S is the co-founder and Chief Science Officer of Kinarm, which commercializes the robotic technology used in the present study.

Poster

PSTR151. Sensorimotor Transformations: Human and Nonhuman Primate

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR151.10/BB11

Topic: D.07. Visual Sensory-Motor Processing

Title: Monocular vision in 3D motion perception provides more accurate visual timing than binocular one

Authors: *O. LEVASHOV;
Brain & Body Develop. Ctr., Da Lat City, Viet Nam

Abstract: To return the tennis ball after your opponent's service we need to obtain a good feeling of visual estimation of time before our kick. This feeling is called “reaction to a moving object” or “timing”. To measure the timing in real 3D we have designed an experimental device that consisted of an inclined gutter with rolling small balls, two sensors, and an electronic timer. In the 1st experiment, we measured the accuracy of timing in “monocular condition” (MC), when the stimulus (ball) rolled from the left to the right along the gutter for crossing the “finish gate”. The task of a subject (S) was to trace visually the movement of the stimulus and to stop the timer at a moment when the ball was passed through the gate. In the 2nd experiment, we measured the accuracy of timing in “binocular condition” (BC), when a gutter was located perpendicular to S’s body, the ball rolled straight towards his eyes. In this configuration, S can use only binocular vision to determine the moment the ball passes through the finish gate. We compared the scatter of timer data in both MC and BC conditions. The measure of timing accuracy was the number of successful attempts inside the time interval {-10 ms + 10 ms}. This interval is a data scatter relative true moment of finish line crossing. A total of 14 Ss participated in both experiments. 12 Ss showed the superiority of MC compared to BC. The results of 2 Ss were similar in both conditions. The results obtained can be explained by a slow velocity of convergent eye movement in BC compared with eye tracking in MC.

Disclosures: O. Levashov: None.

Poster

PSTR151. Sensorimotor Transformations: Human and Nonhuman Primate

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR151.11/BB12

Topic: D.07. Visual Sensory-Motor Processing

Support: NIH Grant R21 NS118055
National Science Foundation Graduate Research Fellowship Program (NSF GRFP)
Rackham Merit Fellowship from the University of Michigan

Title: State-dependent effects of parietal intermittent theta burst stimulation on visuomotor control circuits

Authors: *J. A. DELUISI[1], E. R. GOLDENKOFF[1], T. G. LEE[2], J. A. BRISSENDEN[2], S. F. TAYLOR[3], T. A. POLK[2], M. VESIA[1];
[1]Sch. of Kinesiology, [2]Dept. of Psychology, [3]Dept. of Psychiatry, Univ. of Michigan, Ann Arbor, Ann Arbor, MI

Abstract: Transcranial magnetic stimulation (TMS) can induce changes in cortical excitability and functional connectivity within brain networks. However, the physiological effects of TMS

depend on the activation state at the time of stimulation of the targeted cortical site and its interconnected regions. Our group and others have demonstrated that functional interactions derived from dual-site TMS are context-dependent. For example, the connectivity between the posterior parietal cortex (PPC) and primary motor cortex (M1) shows increased excitability during the planning phase of reaching-to-grasp actions compared to a task-free state (e.g., rest). Yet, the behavioral context during stimulation is often overlooked, despite its potential to contribute to interindividual variability in stimulation outcomes within research and clinical settings. In the current study, we test the hypothesis that inducing specific behavioral states during TMS can selectively enhance the involvement of neuronal populations associated with visuomotor control. Forty-eight healthy adults aged 18-50 were randomly assigned to one of three groups. Participants underwent five daily TMS sessions, during which they received intermittent theta burst stimulation (iTBS) to left PPC to enhance activity in visuomotor control circuits. One group underwent iTBS to the PPC while concurrently performing a grasping task. A second group underwent iTBS to the PPC while in an unconstrained rest state. The third group underwent iTBS to a cortical region outside the grasping network while performing the grasping task concurrently. Participants completed an initial neuroimaging session with resting-state and task-based fMRI. Following the final stimulation session, participants completed an immediate post-TMS neuroimaging session and a one-week follow-up. Visuomotor performance was assessed using a precision force-tracking task inside the scanner, where participants used grip force to match the movement of a cursor to a moving target. Resting-state fMRI analyses measured changes in functional connectivity to the stimulation site from their initial baseline to immediate post-TMS and one-week follow-up. Preliminary results indicate that PPC stimulation affects tracking performance relative to the control site stimulation. Furthermore, initial resting-state and task-based fMRI analyses reveal differential changes in activity patterns due to state-dependent stimulation. These preliminary findings provide insight into the effects of parietal TMS and behavioral state on visuomotor performance and the activity in parietal-frontal circuits associated with action control.

Disclosures: **J.A. Deluisi:** None. **E.R. Goldenkoff:** None. **T.G. Lee:** None. **J.A. Brissenden:** None. **S.F. Taylor:** None. **T.A. Polk:** None. **M. Vesia:** None.

Poster

PSTR151. Sensorimotor Transformations: Human and Nonhuman Primate

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR151.12/BB13

Topic: D.07. Visual Sensory-Motor Processing

Support: Italian Ministry of Research PRIN 20208RB4N9_004

Title: Distinct neural encoding of direction and velocity parameters in parietal and premotor areas during the observation of point light displays grasping actions

Authors: *S. ZICCARELLI[1], A. ERRANTE[1,2], L. FOGASSI[1];
[1]Univ. of Parma, Parma, Italy; [2]Neuroradiology unit, Univ. Hosp. of Parma, Parma, Italy

Abstract: The human ability to process kinematic information embedded in observed actions is essential for understanding others' behavior. Previous studies demonstrated that some action kinematic features may be encoded within the action observation network (AON). However, the neural coding of direction and velocity kinematic parameters in the human brain remains incompletely understood. In the present study we investigated, by means of functional magnetic resonance imaging (fMRI), the neural substrates specifically activated by the direction and velocity of hand grasping actions, which were presented as point light displays (PLDs), thus isolating the action's kinematic features from other informative features. Twenty-three healthy adult participants took part in the event-related fMRI study. They observed videos of PLDs grasping actions executed with two different velocities and four grips in right and left directions. Univariate analysis was employed to identify brain regions differentially recruited by grasping direction and velocity. Our findings show that direction is encoded in posterior visual areas, while velocity recruits MT/V5 and posterior parietal areas. To explore the multivariate pattern of activity within the AON in all experimental conditions, we employed representational similarity analysis (RSA), demonstrating that the activity pattern in posterior intraparietal areas was best explained by the direction model, whereas the pattern in MT/V5 was most closely related to velocity model. Additionally, we employed multi-kernel and multiclass machine learning algorithms to perform multivariate pattern analysis (MVPA). This approach allowed to identify: a) model decoding accuracy of velocity and direction at the network level; b) the crucial role played by MT/V5 in decoding direction, in addition to velocity; c) the role of bilateral sectors of premotor areas, which contribute to velocity decoding. These findings provide compelling evidence that kinematic parameters of hand grasping actions are encoded in both parietal and premotor areas within the AON. They further suggest that parietal areas are primarily involved in coding grasping direction, while MT/V5 and premotor areas in coding grasping velocity, although there are brain areas playing a key role in encoding both parameters. These results could have relevant implications for observational learning strategies based on action kinematics.

Disclosures: S. Ziccarelli: None. **A. Errante:** None. **L. Fogassi:** None.

Poster

PSTR151. Sensorimotor Transformations: Human and Nonhuman Primate

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR151.13/BB14

Topic: D.07. Visual Sensory-Motor Processing

Title: Power Spectral Density Analysis of Kinematic Variables Reveals Age-related Differences in Arm Motion

Authors: *A. A. SHANGHAVI[1], R. PATIL[1], J. STAD[1], G. MAFFEI[2], B. S. DUERSTOCK[1,3], A. B. SERENO[1,4,5];

[1]Weldon Sch. of Biomed. Engin., [2]Sch. of Mechanical Engin., [3]Sch. of Industrial Engin., [4]Dept. of Psychological Sci., Purdue Univ., West Lafayette, IN; [5]Sch. of Med., Indiana Univ., Indianapolis, IN

Abstract: Ageing is associated with changes in physical function, including decline in muscle strength, joint mobility, and cognitive processing. These changes can affect the performance of daily activities, including arm motion. Power spectrum analyses have been used in previous studies to detect abnormal arm motion artifacts such as tremor and rigidity in older as well as neurodegenerative populations. Understanding the differences in arm motion between young and old individuals, using non-invasive sensors, can help improve the detection, evaluation, and design of assistive devices and rehabilitation programs for older adults. This study aimed to identify accurate and reliable changes due to ageing in wrist velocity and acceleration (Postural task) as well as in latency of responses (Pronation-Supination task). Eighteen young (ages 18-20, 7 female) and nine older (ages 49-57, 7 female) adult subjects performed Postural (participants holding their wrists steady) and Pronation-Supination tasks. Wearable inertial measurement units (recording at 100 Hz) placed on both wrists recorded linear acceleration and angular velocity (each in 3 dimensions), which were used to calculate linear velocity and angular acceleration (12 kinematic variables total) and response latency. Using the total power of spectral densities of the 12 wrist kinematic variables, a linear Support Vector Machine classifier successfully differentiated between young and old populations in the Postural task with up to 90.9% accuracy, 100.0% specificity and 90.0% sensitivity. Results indicate that the best differentiation was found between 4-8 Hz and 9-13 Hz. Analysis of velocity in the roll-axis in the Pronation-Supination task revealed that older adults had a 68ms slowing in response latency compared to younger adults. We show that lightweight wrist sensors provide detectable, accurate and reliable measures of age-related arm movement changes. Future work will focus on extending these methods to determine distinguishing features in arm motion between young, old and Parkinson's Disease populations.

Disclosures: A.A. Shanghavi: None. **R. Patil:** None. **J. Stad:** None. **G. Maffei:** None. **B.S. Duerstock:** None. **A.B. Sereno:** None.

Poster

PSTR151. Sensorimotor Transformations: Human and Nonhuman Primate

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR151.14/BB16

Topic: D.07. Visual Sensory-Motor Processing

Support: Canadian Institutes of Health Research (CIHR)
Natural Sciences and Engineering Research Council (NSERC)
Vision: Science to Applications (VISTA) program

Title: Hierarchical Functional Modularity of Brain Networks for Egocentric and Allocentric Memory-guided Reaching

Authors: *L. MUSA[1,2,3], A. GHADERI[3], Y. CHEN[3], J. CRAWFORD[3,2,4,5];
[2]Psychology, [3]Ctr. for Vision Res., [4]Kinesiology, [5]Biol., [1]York Univ., North York, ON, Canada

Abstract: The brain can encode targets for reaching in egocentric and/or allocentric reference frames (Byrne and Crawford 2010). The differences in the cortical activation of these two representations has been described (Chen et al., 2014; Neggers et al., 2006). For example, Chen et al. (2014) identified egocentric directional selectivity in dorsal brain areas (the parieto-frontal cortex) versus landmark-centered directional selectivity in ventral brain areas (inferior temporal gyrus and inferior occipital gyrus) during a delayed reach task. However, differences in the functional organization of brain networks have not been studied. Here, we performed a secondary analysis of the event-related fMRI task from Chen et al. (2014), to distinguish human brain networks involved in egocentric versus allocentric spatial representation of reach goals. Based on their previous univariate analysis we expected that the functional brain networks will differ, with increased hubness in ventral brain regions in the allocentric task. The paradigm consisted of three tasks with identical stimulus display but different instructions: egocentric reach (remember absolute target location), allocentric reach (remember target location relative to a visual landmark), and a nonspatial control, color report (report color of target). We performed a graph theoretical analysis (GTA) on time series data recorded during the entire trial, contrasting egocentric and allocentric data versus controls and each other. The fractal organization of the network modules was determined using agglomerative and divisive clustering approaches. Modularity maximization and consensus partitioning were used to identify the scale of modules that best delineate egocentric and allocentric brain networks. A comparison with resting state network parcellations showed poor overlap, with decreased modularity in resting state networks. Instead, the data were largely segregated into dorsal and ventral modules, with similar organization in egocentric vs. allocentric trials. However, the egocentric network demonstrated significantly stronger modularity in parietal modules. Brain regions that were important for connecting different modules in the allocentric task were found ventrally in the parahippocampal cortex and inferior extrastriate visual cortex, whereas in the egocentric task those hubs were found in the superior extrastriate visual cortex and the interparietal lobule. We conclude that egocentric versus landmark-guided reach utilizes largely overlapping cortical networks, but with significant differences consistent with a dorsal-ventral specialization.

Disclosures: L. Musa: None. **A. Ghaderi:** None. **Y. Chen:** None. **J. Crawford:** None.

Poster

PSTR151. Sensorimotor Transformations: Human and Nonhuman Primate

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR151.15/BB17

Topic: D.07. Visual Sensory-Motor Processing

Support: NIH Grant UL TR002001
NIH Grant KL2 TR001999
NIH Grant R01NSO89069

Title: Parietal lesions disrupt manipulable object representations in the ventral visual pathway.

Authors: *F. GARCEA[1], E. STRAWDERMAN[2], S. MEYERS[3], K. WALTER[2], T. SCHMIDT[2], W. H. PILCHER[2], B. MAHON[2];
[1]Neurosci., [2]Neurosurg., [3]Imaging Sci., Univ. of Rochester, Rochester, NY

Abstract: The left inferior parietal lobule (LIPL) is a key substrate for object grasping and manipulation. The goal of this project was to test the hypothesis that lesions to parietal areas sensitive to object-directed grasping would disrupt neural responses for manipulable objects in ventral temporal cortex (VTC). Evidence consistent with this hypothesis would indicate that category-specificity for manipulable objects in the VTC is shaped in part from inputs from LIPL regions that support object grasping and manipulation. To test this hypothesis, we used a novel method—Voxelwise Lesion-Activity Mapping (VLAM)—to investigate how lesions to anatomically remote regions modulate neural responses for manipulable objects in the VTC. One hundred seven participants with an acquired brain lesion took part in an fMRI experiment to localize neural responses for manipulable objects in the VTC. Each participant's lesion was hand-drawn on their T1 anatomical scan and normalized into MNI space; we then investigated the lesion sites associated with reduced neural responses in the VTC. We found that lesions to the left anterior intraparietal sulcus (aIPS), a region that supports hand-shaping during object grasping, and the left supramarginal gyrus (SMG), a region that supports the retrieval of manipulation knowledge, were associated with reduced neural responses for manipulable objects in the left VTC. Control analyses demonstrated that neural responses to place stimuli in the left VTC were unaffected by lesions to the left aIPS or left SMG, suggesting a domain-specific parietal modulation of responses in the left VTC. Using a separate dataset from 55 neurotypical controls who participated in the same fMRI experiment, we confirmed that neural responses for manipulable object stimuli were maximal in the portion of the left aIPS and left SMG identified in the VLAM analysis. Our findings provide causal lesion evidence that neural responses in the left VTC are selectively modulated by parietal actions areas in the left aIPS and left SMG. More broadly, these data suggest that category-specific neural responses for manipulable objects in the VTC emerge, in part, due to inputs that come by way of parietal regions that support object-directed actions.

Disclosures: F. Garcea: None. **E. Strawderman:** None. **S. Meyers:** None. **K. Walter:** None. **T. Schmidt:** None. **W.H. Pilcher:** None. **B. Mahon:** None.

Poster

PSTR151. Sensorimotor Transformations: Human and Nonhuman Primate

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR151.16/BB18

Topic: D.07. Visual Sensory-Motor Processing

Support: VISTA (Science to Application)
Discovery NSERC Grant

Title: Shape sensitivity in the dorsal pathway is dissociable from attentional processes

Authors: *Y. GOLDSTEIN MARCUSOHN, E. FREUD;
York Univ., Toronto, ON, Canada

Abstract: The two visual pathways model insinuates that different visual behaviors are supported through two separate cortical systems: the ventral pathway and the dorsal pathway. The ventral pathway promotes visual recognition, allowing us to identify and categorize objects, while the dorsal pathway supports visually guided actions. Notably, however, accumulating evidence demonstrates that the dorsal pathway also plays a role in shape processing and object perception, challenging the dichotomy of the functional dissociation. However, the nature of object representations in the dorsal pathway is yet to be determined. Several recent findings pointed out that these representations are particularly tuned to attentional manipulations and tasks demands. Hence, an outstanding question is to what extent shape sensitivity in regions of the dorsal pathway are dissociable from attentional mechanisms. To address this question, we conducted an fMRI experiment. Human participants viewed novel objects that were presented in different levels of scrambling to manipulate the availability of shape information. Participants focused their attention on either the color of the objects or the background. We employed univariate and multivariate analyses to map the large-scale organization of shape sensitivity across the two visual pathways. The sensitivity to shape information increased from the early visual cortex to the extrastriate cortex in both pathways and then slightly decreased in more anterior regions of each pathway. Critically, while the attentional manipulation did modulate shape-selective responses in regions of both pathways, the large-scale shape sensitivity maps were identified at the individual level, regardless of the participants' attentional focus. Together, these findings provide support for the notion that shape processing relies on a distributed network of cortical regions across the visual pathways, and this process appears to be dissociable of attentional mechanisms.

Disclosures: Y. Goldstein Marcusohn: None. **E. Freud:** None.

Poster

PSTR151. Sensorimotor Transformations: Human and Nonhuman Primate

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR151.17/BB19

Topic: D.07. Visual Sensory-Motor Processing

Support: NIH Grant 1R01EY030854

Title: Understanding the interaction of attention and working memory load in visual object representation in human occipito-temporal and posterior parietal cortices

Authors: *Y. XU;
Yale Univ., New Haven, CT

Abstract: Attention and working memory load have both been shown to impact visual object representation in the human brain. How do these two factors jointly determine visual object representation in the human brain? In this fMRI study, 12 human observers viewed blocks of object images, with each block containing a sequential presentation of different exemplars from the same object category. A total of four different object categories were show and they were cars, cats, faces and houses. Each object image also contained a set of transparent colored dots superimposed on the object images. All the dots for a given image were of the same color. Observers were asked to attend to either the shape of the object or the color of the dots and performed either a 1-back or a 2-back repetition detection task on the attended feature. Object category decoding was then examined across the occipito-temporal cortex (OTC) and posterior parietal cortex (PPC). While the object representational geometry was determined predominantly by object category in OTC, in higher PPC regions, it was determined by attention, load, and, to a minor extent, object category. Interestingly, attention and load appears to form two near orthogonal axes in PPC in the representational space. Cross-decoding further revealed stronger attention independent load decoding and load independent attention decoding in PPC than OTC regions. Together, these results delineate for the first time how objects, attention and load jointly determine visual object representation across different areas in human OTC and PPC.

Disclosures: Y. Xu: None.

Poster

PSTR151. Sensorimotor Transformations: Human and Nonhuman Primate

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR151.18/BB20

Topic: D.07. Visual Sensory-Motor Processing

Support: UG1EY032039
R01EY015545
Tianqiao and Chrissy Chen Brain-machine Interface Center at Caltech
Boswell Foundation
Swartz Foundation

Title: Abstract spatial information in human PPC

Authors: *J. A. GAMEZ[1], T. AFLALO[1], K. KADLEC[1], C. GUAN[1], X. ZOU[1], K. PEJSA[1], E. ROSARIO[2], N. POURATIAN[3], A. BARI[3], R. A. ANDERSEN[1];
[1]Div. of Biol. and Biol. Engin., Caltech, Pasadena, CA; [2]Casa Colina Centers for Rehabil., Pomona, CA; [3]Neurosurg., UCLA, Los Angeles, CA

Abstract: The human posterior parietal cortex (PPC) has been implicated in functions such as motor planning, spatial attention, decision-making, and semantic representations. These functions suggest a complex role of this brain area in the processing of sensory and motor information. Furthermore, damage to PPC has been associated with dysfunctions in the

processing of spatial information. Therefore, a better understanding of how PPC processes spatial information is important for the design of brain-machine interfaces. Using a cognitive spatial task, where the participant had to decide the spatial relationship of an object with respect to a landmark, we previously found that human PPC encodes spatial information in allocentric and egocentric reference frames during the early stimulus presentation phase of the task. This spatial information becomes an abstract representation, independent of the original reference frame, during the response planning and execution phases of the task. The next question that we wanted to answer was, does PPC represent other abstract variables during the motor planning process? To answer this question, we recorded from the anterior part of the superior parietal lobule (SPL) in PPC and the hand knob in the motor cortex (MC) of one human tetraplegic participant implanted with one 96-channel Blackrock NeuroPort array in each of these brain areas as part of a brain-machine interface clinical trial. The participant performed a variation of the cognitive spatial task described above. In this new version, after making his decision on the spatial relationship between the object and the landmark during the stimulus presentation phase, the participant had to use symbolic information (left/right arrow) or a spatial color code (blue=left / red=right) to plan and execute a saccade to a target that represented his decision. As in the previous version of the cognitive spatial task, we found that MC did not encode the participant decision, while SPL encoded the spatial decision during the stimulus presentation and motor planning and execution phases. However, during the saccade planning and execution phases, SPL encoded information about the spatial decision for the symbolic version of the task stronger than for the color version. These results show that SPL preferentially encodes the spatial decision related to symbolic information over a color representation.

Disclosures: **J.A. Gamez:** None. **T. Aflalo:** None. **K. Kadlec:** None. **C. Guan:** None. **X. Zou:** None. **K. Pejsa:** None. **E. Rosario:** None. **N. Pouratian:** None. **A. Bari:** None. **R.A. Andersen:** None.

Poster

PSTR151. Sensorimotor Transformations: Human and Nonhuman Primate

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR151.19/BB21

Topic: D.07. Visual Sensory-Motor Processing

Title: The prevalence of ipsilateral silent periods as a measure of interhemispheric inhibition during mirror illusion

Authors: *C. M. SMITH[1], S. M. REESE[1], J. E. SHIELDS[1], A. A. OLMOS[1], C. C. VOSKUIL[2], X. YE[3], M. S. STOCK[4], J. C. CARR[2], J. M. DEFREITAS[1];
[1]Oklahoma State Univ., Stillwater, OK; [2]Texas Chirstian Univ., Fort Worth, TX; [3]Univ. of Hartford, West Hartford, CT; [4]Univ. of Central Flordida, Orlando, FL

Abstract: The prevalence of ipsilateral silent periods as a measure of interhemispheric inhibition during mirror illusions

AuthorsClaire M. Smith[1], Shawn M. Reese[1], JoCarol E. Shields[1], Alex A. Olmos[1], Caleb C. Voskuil[2], Xin Ye[3], Matt S. Stock[4], Joshua C. Carr[2], Jason M. DeFreitas[1] [1]Oklahoma State University, Stillwater, Oklahoma; [2]Texas Christian University, Fort Worth, Texas; [3]University of Hartford, West Hartford Connecticut; [4]University of Central Florida, Orlando, Florida.
Mirror therapy is a commonly used therapeutic intervention in physical therapy rehabilitation clinics for patients suffering from unilateral neural disorders, such as loss of limb (phantom limb pain), stroke, or crushing injuries. Ipsilateral silent periods (iSP), a measure of interhemispheric inhibition from transcranial magnetic stimulation (TMS) might be able to provide useful information regarding the interhemispheric communication during mirror therapy. However, iSPs are somewhat unpredictable, and are not consistently present with each stimulation. It is possible though, that the frequency of iSPs might, in turn, be a useful metric. **Purpose**: The purpose of this study was to quantify whether the frequency of ipsilateral silent periods would be sensitive to various contraction types and mirror illusions. **Methods:** Twenty-nine healthy subjects (18-35 yrs) participated in this study. A figure-eight TMS coil was used to stimulate the hand area of the right primary motor cortex during dynamic and isometric contractions of the right hand under various visual conditions. Participants were instructed to fix their gaze in the same location for three visual conditions: No mirror, bilateral mirror, and unilateral mirror illusion. These conditions are believed to alter interhemispheric communication. **Results:** A 2-way repeated measures ANOVA showed neither a contraction × condition interaction ($p = 0.447$), nor significant main effects for contraction or mirror condition ($p = 0.963$, $p = 0.162$, respectively). While the prevalence of iSPs ranged from 0% to 100% of stimulation trials across individuals, the group means were fairly homogenous across conditions, all falling within a range of 45.9% to 54.5%. **Conclusion:** While there is previous evidence that the duration of ipsilateral silent periods may be a useful and sensitive measure of interhemispheric inhibition across various conditions, it does not appear that the prevalence, or frequency, of ipsilateral silent periods is as valuable.

Disclosures: C.M. Smith: None. **S.M. Reese:** None. **J.E. Shields:** None. **A.A. Olmos:** None. **C.C. Voskuil:** None. **X. Ye:** None. **M.S. Stock:** None. **J.C. Carr:** None. **J.M. DeFreitas:** None.

Poster

PSTR151. Sensorimotor Transformations: Human and Nonhuman Primate

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR151.20/BB22

Topic: D.07. Visual Sensory-Motor Processing

Title: The Effect of Mirror Illusion and Contraction Type on Interhemispheric Inhibition

Authors: *S. M. REESE[1], C. M. SMITH[1], J. SHIELDS[1], A. A. OLMOS[1], C. C. VOSKUIL[2], X. YE[3], M. S. STOCK[4], J. C. CARR[2], J. M. DEFREITAS[1];
[1]Oklahoma State Univ., Stillwater, OK; [2]Texas Christian Univ., Fort Worth, TX; [3]Univ. of Hartford, Harford, CT; [4]Univ. of Central Florida, Orlando, FL

Abstract: Mirror illusions are utilized in the physical rehabilitation of impaired limbs due to injury and stroke. The premise is to use contractions of the unaffected limb combined with a visual mirror illusion to stimulate the affected limb's associated motor cortex and peripheral neural pathways without actual physical movement of the limb. Transcranial magnetic stimulation (TMS) allows us to quantify the magnitude of interhemispheric communication under various conditions, and may allow us to assess the interhemispheric effects of mirror illusions. **PURPOSE:** To quantify the effects of different contraction types and mirror illusion conditions that vary the degree of visual input on interhemispheric inhibition. **METHODS:** Thirteen healthy individuals have completed this study. A figure-eight TMS coil was used to stimulate the hand region of the right primary motor cortex. Ipsilateral Silent Period (iSP) was used as a measure of interhemispheric inhibition, and was measured utilizing surface electromyography placed over the right first dorsal interosseous (FDI) muscle during a series of dynamic and isometric contractions. A series of 10 TMS stimulations were conducted at 140% of resting motor threshold with an interstimulus interval of 5-10 seconds during dynamic and isometric abduction of the FDI muscle at 40% of maximum under 3 mirror illusion conditions. Participants were instructed to fix their gaze in the same location for each of the 3 mirror conditions which included: No mirror, Bilateral, and unilateral mirror illusion. A 2 way contraction × condition repeated measures ANOVA was used to assess the iSP's. **RESULTS:** The ANOVA showed there was no contraction × condition interaction ($p = 0.899$), but there was a main effect for contraction type ($p < 0.001$). The isometric contractions demonstrated a mean iSP that was 5.44ms longer than during dynamic contractions (i.e. greater inhibition), regardless of the mirror condition. **CONCLUSION:** The degree of interhemispheric inhibition may be affected by contraction type with an increase in inhibition when utilizing isometric contractions compared to dynamic contractions. The results of this study show no effect of mirror illusion on interhemispheric inhibition, which perhaps suggests other possible transcallosal mechanisms should be explored, such as interhemispheric facilitation. This research demonstrates contraction type should be considered when prescribing mirror therapy to patients and may provide insight into rehabilitation programming.

Disclosures: S.M. Reese: None. **C.M. Smith:** None. **J. Shields:** None. **A.A. Olmos:** None. **C.C. Voskuil:** None. **X. Ye:** None. **M.S. Stock:** None. **J.C. Carr:** None. **J.M. DeFreitas:** None.

Poster

PSTR151. Sensorimotor Transformations: Human and Nonhuman Primate

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR151.21/BB23

Topic: D.07. Visual Sensory-Motor Processing

Support: National Institute of Mental Health Intramural Research Program ZIA-MH-002909

Title: Understanding Object Affordances Using Verb Usage Patterns and Behavioral Experiments

Authors: *M. VAZIRI PASHKAM[1], N. PALLIS-HASSANI[2], A. MIREBRAHIMI TAFRESHI[3], A. ZOROUFI[4], K. LAM[2], F. PEREIRA[5], C. BAKER[5];
[1]Lab. of Brain and Cognition, [2]Natl. Inst. of Mental Hlth., Bethesda, MD; [3]Western Univ., London, ON, Canada; [4]MIT, Cambridge, MA; [5]Natl. Institue of Mental Hlth., Bethesda, MD

Abstract: When we see objects, we immediately know not only what they are, but also how to interact with them. Yet little research has been performed to understand what information people glean from objects about the interactions they support. The first step towards revealing the neural correlates of such object affordances is to understand people's knowledge of an object's action associations. Here, we first used language as a means to tap into humans' knowledge of what actions can be performed with an object. Using a large database of ~1850 object categories (THINGS database) and ~5000 verbs, we identified applications of each verb to each object in a large text corpus. We then used this data to embed each object in a space where dimensions correspond to verbs that apply to similar objects. We showed, in behavioral experiments, that these extracted embedding dimensions are meaningful to human observers. Next, to reveal people's understanding of potential actions towards objects, we conducted online behavioral experiments in which we presented images of individual objects from the THINGS database and asked people action related questions: if they can pick up the objects with one or two hands, if the object is strongly associated with a specific hand action, what grasp type do they use to pick up the object, and what body parts do they use to interact with the objects. For 86% of the objects in the database, participants indicated that they could pick them up with one or two hands. Many objects, including both tool and non-tool items, had a strong action association. Although hand was the most common body part implicated, other body parts were also reported to be heavily involved in interacting with objects. Lastly, the tripod grasp was the most commonly reported grasp type for picking up the objects, with the pinch and cylindrical grasps coming second and third. Together, these results indicate strong object-action associations evident in both text corpora and in people's reports from viewing pictures of objects. They uncover the richness of object interactions and argue for moving beyond simple hand grasps and beyond the specific category of tools in future behavioral and neuroscientific experiments.

Disclosures: M. Vaziri Pashkam: None. **N. Pallis-Hassani:** None. **A. Mirebrahimi Tafreshi:** None. **A. Zoroufi:** None. **K. Lam:** None. **F. Pereira:** None. **C. Baker:** None.

Poster

PSTR152. Cross-Modal Processing in Humans

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR152.01/BB24

Topic: D.08. Multisensory Integration

Support: Undergraduate Research in Science and Technology Grant

Title: Non-conscious multi-sensory integration revealed in the ventriloquist effect

Authors: *E. TURKOVICH, J. SAMAHA, S. SANKARAN, W. DOU;
Psychology, Univ. of California, Santa Cruz, Santa Cruz, CA

Abstract: The degree to which information from distinct sensory modalities can interact in the absence of conscious awareness remains controversial. According to the Global Workspace Theory (GWT), unconscious sensory information remains relatively confined to the sensory cortex and should not interact with other modalities until it is broadcast into the (conscious) global workspace comprising frontal-parietal areas. The ventriloquist effect refers to the misperception of a sound location towards that of a concurrent visual stimulus, such as perceiving the voice of a ventriloquist as coming from the moving dummy. Here, we used visual masking to render the location of a brief flash stimulus unconscious while participants performed a sound localization task. Preliminary results indicate that, despite being at chance levels in discriminating the flash location, participants were nevertheless biased to localize sounds towards the unconscious flash locations. In line with recent findings from Delong et al (2018), this suggests that consciousness is not required for the integration of signals originating in distinct sensory modalities. Further, preliminary ERP results suggest that audiovisual integration may be happening as early as 200ms, preceding the suggested timeline for conscious ignition, theorized to be around 300ms by the brain-based version of GWT: Global Workspace Dynamics. These combined behavioral and EEG results might prompt revision of certain features of the GWT.

Disclosures: E. Turkovich: None. **J. Samaha:** None. **S. Sankaran:** None. **W. Dou:** None.

Poster

PSTR152. Cross-Modal Processing in Humans

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR152.02/BB25

Topic: D.08. Multisensory Integration

Title: Concurrent headshake and postural training using virtually reality improves balance and modifies muscle activation: Preliminary data

Authors: *K. O. APPIAH-KUBI[1], H. SHATTUCK[1], J. BICKNELL[1], E. LAING[2], J. VANLANDEGHEM[3], D. SENARATHNA[4], M. IMTIAZ[5];
[1]Physical therapy, [2]Mechanical & Aerospace Engin., [3]Biol., [4]Mathematics, [5]Electrical and Computer Engin., Clarkson Univ., Potsdam, NY

Abstract: Background: Vestibular rehabilitation has been shown to improve postural balance by influencing sensory reweighting. Several studies use gaze stabilization exercises (such as headshake activities) and balance training (such as weight shifting) at separate times of the training sessions to improve vestibular function. However, no study has employed a concurrent headshake and weight shift training using virtual reality (Concurrent VR HS-WST) to enhance

the vestibular system and simultaneously improve postural balance. **Purpose:** To assess changes in postural balance and vestibular-motor responses after virtual reality Concurrent HS-WST. **Methods:** Seven healthy young adults (age = 21.0±1.4years; height = 1.7±0.1m) participated in this crossover design study. They began as controls for six consecutive days. Four days later, the participants then performed the Concurrent VR HS-WST for six consecutive days. The participants donned an Oculus Quest 2 headset in a standing position on the floor or Airex foam. They followed the virtual environment (UpRight VR, LLC, Temple University) which instructed them to shift their center of mass (COM) as quickly to accurately reach a visual target that surrounded their COM (every four seconds) in eight possible directions. While participants performed the weight shift, in some of the exercises, they concurrently rotated the head in a horizontal rhythmic pattern (+30°) synchronously to a metronome (80-120 beats/min). Each day's session comprised 18 exercises, each lasting for one minute. Pre- and post- balance, vestibular and muscle activation assessments were performed during the control and training periods. Significance was set at $p \leq 0.05$. **Results:** The modified Clinical Test of Sensory Interaction on Balance showed an appreciable reduction in sway equilibrium scores in the foam eyes open, foam eyes closed and composite scores of 0.083±0.047 (p = 0.87), 0.150±0.031 (p = 0.31) and 0.067±0.0162 (p = 0.21), respectively. The control period showed relatively smaller or no changes. During the ramp-up (i.e., throwing participants backwards) and down (i.e., throwing participants forwards) perturbation trials, all the postural muscles increased in their electromyography (EMG) amplitude, except for biceps femoris which exhibited a decrease in muscle activation (ramp up = 20.93±15.34 mv, p = 0.05; ramp down = 16.16±21.18 mv, p = 0.51). **Conclusion:** The preliminary results of this study lean toward the ability of the Concurrent VR HS-WST to improve postural balance during more challenging postural behaviors when the standing surface is unreliable and sometimes when vision is limited.

Disclosures: K.O. Appiah-Kubi: None. **H. Shattuck:** None. **J. Bicknell:** None. **E. Laing:** None. **J. VanLandeghem:** None. **D. Senarathna:** None. **M. Imtiaz:** None.

Poster

PSTR152. Cross-Modal Processing in Humans

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR152.03/Web Only

Topic: D.08. Multisensory Integration

Title: Analysis of functional network connectivity dynamics and meta-state metrics of intravesical cold stimulation

Authors: *B. JARRAHI;
Stanford Univ., Palo Alto, CA

Abstract: The primary afferent fibers transmit physiological information from the body's viscera to the central nervous system. The smaller-diameter, unmyelinated C-fibers are responsible for transmitting interoceptive signals related to infection, thermal, or chemical stimuli. The current

study aims to investigate the impact of cold stimulation of the bladder (intravesical) tissue, which activates C-fibers, on the dynamic connectivity of the brain network architecture. We used group ICA to decompose BOLD fMRI data of a resting-state condition and a cold-bladder stimulation task, collected from 13 healthy females on a 3.0 T scanner. We estimated the time-varying functional network connectivity by calculating sliding time-window correlations and performing k-means clustering of the windowed correlation matrices. To assess differences in dynamic functional network connectivity (dFNC) between the resting-state and bladder cold stimulation conditions, we used the median of dFNC correlations across the windows and evaluated variations in dynamic correlation patterns using a paired t-test. Furthermore, we employed the meta-state dynamics method by reducing the number of windowed FNC correlations to four components/clusters (based on the elbow criterion) using the following methods: k-means, PCA, temporal ICA, and spatial ICA (utilizing ICA by entropy bound minimization method; ICA-EBM). The dFNC correlations were factorized into continuous loading coefficients and discretized using quartile discretization. Paired t-tests were then performed to compare the following metrics: the number of meta-states, changes in meta-states, meta-state span (i.e., the maximum L1 distance between states for each subject), and the total distance traveled in the n-dimensional state space. The results revealed major changes in several dFNC correlations due to intravesical cold stimulation, particularly showing lower dFNC among brainstem, cerebral, prefrontal, and visual networks at an exploratory uncorrected p-value of 0.001. The meta-state analyses indicated a trend towards a longer total traveled distance during intravesical cold stimulation compared to the resting-state condition (temporal ICA $p = 0.06$). These preliminary results suggest noteworthy effects of intravesical cold stimulation on the dynamics of functional connectivity. If confirmed with larger sample sizes, these findings provide further evidence for the impact of intravesical cold stimulation on the reorganization of the brain's functional network connectivity, highlighting the involvement of key brain regions such as the cerebellar/brainstem and prefrontal networks.

Disclosures: B. Jarrahi: None.

Poster

PSTR152. Cross-Modal Processing in Humans

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR152.04/CC1

Topic: D.08. Multisensory Integration

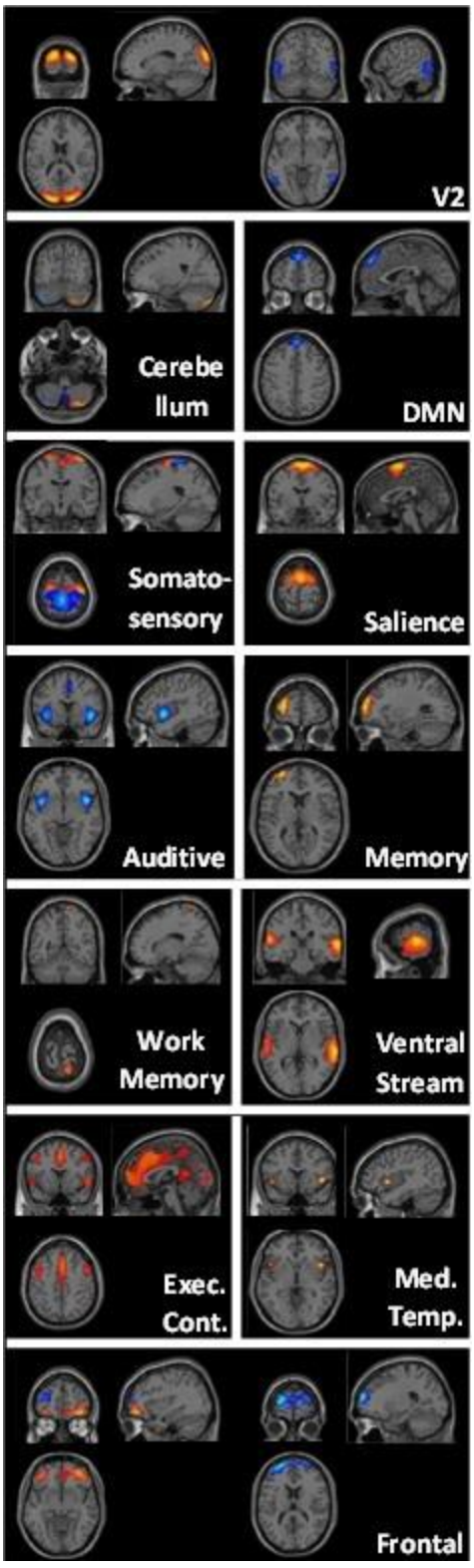
Support: FONCICYT/37/2016, I000/667/2016
Medical Research Council United Kingdom - CONACyT Research Partnerships, Call 2015, Newton Fund

Title: Cortical plasticity of resting state networks in obese vs. normo-weight children.

Authors: *B. DE CELIS ALONSO[1], S. HIDALGO TOBÓN[2], M. ANTONIO DE LA ROSA[1], P. DIES SUÁREZ[2], M. KLUNDER KLUNDER[3], B. LÓPEZ MARTÍNEZ[4], E. BARRAGÁN

LÓPEZ[5], A. MIRANDA LORA[6], P.-W. SO[7];
[1]Fac. of Physical and Mathematical Sci., Benemerita Univ. Autonoma De Puebla, Puebla, Mexico; [2]Imagenology Dept., [3]Dept. of community health, [4]Subdirección de Servicios Auxiliares y de Diagnóstico, [5]Neurol. Dept., [6]Med. Res. unit based on evidence, Hosp. Infantil de México Federico Gómez, Mexico City, Mexico; [7]Dept. of Neuroimaging, King´s Col. London, London, United Kingdom

Abstract: Obesity and its associated comorbidities represent a health risk very relevant to children, as it can affect their cognitive development. Our aim was **t**o point out the differences in brain areas recruited and Functional Connectivity (FC) during resting state network (RSNWs) analysis of an Obese (OB) and a Normo-weight (NW) infant cohorts. 126 male children with ages within 7 and 10, were subdivided into a NW group and an OB group according to their BMI. The grouped RSNWs for each cohort was then calculated. Differences between the RSNWs regions recruited as well as in FC within and with other RSNWs were calculated. **Figure 1** visually presents the results of a T-test comparison of OB vs. NW form for each RSNW. positive voxels (OB>NW) was compared vs. negative voxels (NW>OB), and there was a relationship of 3.05 to 1 (7852 voxels vs. 2574). OB volunteers expanded their RSNWs into adjacent brain regions more than NW volunteers. On top of that, FC within and with other RSNWs and ROI was in general smaller for OB vs. NW. These results mimic cortical plasticity studies in human and animal models. In these works, BOLD activity from a single spared whisker in a rat´s snout, after a few days invaded/recruited surrounding areas of the brain. This was done to compensate for the lack of input that the animal had under these conditions. Could OB humans be doing the same trying to recruit more inputs (motor, auditive, somatosensorial, etc.), so the different functions of day-to-day life can be performed better? Furthermore, these same studies showed through electrophysiology recordings, that in recruited regions spiking activity was less coherent and synchronous. Could this be the reason for reduced FC within and with other RSNS? Still, it is worth considering that the results presented here are resting states, which correspond to low frequency BOLD fluctuations as well as GIFT maps which can be seen as covariance maps of the filtered BOLD signals. This is in contrast with pure BOLD signals obtained from whisker stimulation, which is what was used in the papers of cortical plasticity.



Disclosures: B. De Celis Alonso: None. **S. Hidalgo Tobón:** None. **M. Antonio de la Rosa:** None. **P. Dies Suárez:** None. **M. Klunder Klunder:** None. **B. López Martínez:** None. **E. Barragán López:** None. **A. Miranda Lora:** None. **P. So:** None.

Poster

PSTR152. Cross-Modal Processing in Humans

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR152.05/CC2

Topic: D.08. Multisensory Integration

Support: R00DC013828
1R01DC020717
R01NS094399
NIH NIDCD R01 DC020717

Title: Visual speech increases the decodability of phonemes in auditory cortex

Authors: *A. MAJUMDAR[1], C. CAO[1], W. C. STACEY[2], D. BRANG[3];
[2]Neurol., [3]Psychology, [1]Univ. of Michigan, Ann Arbor, Ann Arbor, MI

Abstract: During the perception of audiovisual speech, listeners use visual speech information (e.g., lipreading) to improve auditory speech processes. Silent lipreading has been shown to modulate activity in the auditory cortex, and we recently showed that the initial consonants from lipread words could be decoded from auditory areas. However, it remains unclear whether lipread signals target and improve the representations of specific phoneme-tuned neuronal populations. To test this hypothesis, we recorded intracranial EEG (iEEG) data from patients with intractable epilepsy during a word identification task. We selected 16 words that were matched to 4 initial phonemes (/m, n, b, g/) with 4 trailing diphones (/ih-l, ey-t, ae-sh, uw-n/) and presented these words in three conditions: audio-only, visual-only, and audiovisual. To explore how visual speech modulates the tuning of phoneme representations in the auditory cortex, we trained SVM classifiers to decode word identity information using activity from superior temporal gyrus electrodes. In the visual-only condition, both the initial consonants and middle phonemes could be significantly classified from activity in auditory electrodes, demonstrating that the auditory system encodes lipreading information. Moreover, audiovisual speech showed higher decoding accuracy compared to auditory-only speech for both the initial consonants and middle phonemes. We interpret these results as being consistent with the hypothesis that visual speech targets matching phoneme-tuned neuronal populations to increase the precision of their neuronal representations, and that this audiovisual transformation occurs throughout the time course of a word, instead of just for the initial phoneme or consonant.

Disclosures: A. Majumdar: None. **C. Cao:** None. **W.C. Stacey:** None. **D. Brang:** None.

Poster

PSTR152. Cross-Modal Processing in Humans

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR152.06/CC3

Topic: D.08. Multisensory Integration

Title: Individual Differences in Cybersickness Development: The Role of Sensory Reweighting in VR

Authors: *S. IZADI SOKHTABANDANI[1], M. BARNETT-COWAN[2];
[2]Univ. of Waterloo, [1]Univ. of Waterloo, Waterloo, ON, Canada

Abstract: Virtual Reality (VR) usage has grown rapidly, bringing into focus the need to understand the factors influencing cybersickness, a condition resulting from immersive VR and augmented reality applications, to enhance user experiences (McCauley & Sharkey, 1993). Susceptibility to cybersickness varies among individuals, necessitating investigations into underlying mechanisms. Building on our lab's recent work (Chung & Barnett-Cowan, 2023), which indicated a relationship between sensory reweighting, particularly the subjective visual vertical (SVV), and cybersickness susceptibility, our current research expands upon the understanding of sensory reweighting using both SVV and the Oriented CHAracter Recognition Task (OCHART), which gauges the perceptual upright (PU). We hypothesized that cybersickness severity is linked to the central nervous system's short-term neural plasticity, with the system reweighting multisensory orientation information to adjust to conflicting inputs between the physical and VR environments. Results to date substantiate that bodily, gravitational, and visual orientation information significantly influence both SVV and PU, reinforcing prior research findings. Importantly, our study shows that high-intensity VR exposure reduces the influence of visual information on orientation perception, suggestive of a down-weighting of visual cues. Additionally, we observed that individuals less prone to cybersickness typically demonstrate greater adaptation in their reliance on visual information. These findings contribute to the understanding of VR aftereffects on sensory perception, hinting at a potential link between cybersickness susceptibility and sensory reweighting. Our continuing research aims to gather more data and utilize a vector sum model to further probe the impacts of the discussed cues on SVV and PU. By comprehending these interactions, we intend to discern individual differences in cybersickness development, enabling the creation of tailored mitigation strategies. This knowledge will enhance VR experiences, fostering greater accessibility and comfort for a wider user demographic.

Disclosures: S. Izadi Sokhtabandani: None. **M. Barnett-Cowan:** None.

Poster

PSTR152. Cross-Modal Processing in Humans

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR152.07/CC4

Topic: D.08. Multisensory Integration

Support: NIH Grant R01NS065395
NIH Grant U01NS113339

Title: Synthetic Faces Improve the Intelligibility of Noisy Speech, But Not As Much As Real Faces

Authors: *Y. YU[1], A. LADO[1], Y. ZHANG[2], J. MAGNOTTI[1], M. S. BEAUCHAMP[1];
[1]Univ. of Pennsylvania, Philadelphia, PA; [2]Baylor Col. of Med., Houston, TX

Abstract: It has long been known that seeing the face of the talker improves the intelligibility of noisy auditory speech. Advances in computer graphics have made encountering synthetic faces an everyday occurrence, but little is known about whether synthetic faces improve speech intelligibility. To address this knowledge gap, audiovisual recordings were created from two human talkers (one male, one female), each speaking 30 different words. Pink noise was added to the voices at a signal-to-noise ratio of -12 dB. Commercial software from JALI Research was used to animate two rigged facial 3D models (one male and female) to create synthetic faces lip synced to the gender-matched voice. Three formats of each word were created: noisy auditory-only (An); noisy audiovisual with a real face (AnR) and noisy audiovisual with a synthetic face (AnS). Thirteen participants were presented with noisy words using the Amazon Mechanical Turk online testing service. Participants viewed a video introduction to the study and then answered the prompt "Type the word" following presentation of each word. The phonetic composition of each typed response was scored for accuracy based on the Carnegie-Mellon pronunciation dictionary (*e.g.*, a response of *wyatt* for *quiet* scored 80%). Each participant was presented with each word only once, in a single format, to prevent any learning effects. Across participants, every word was presented in every format. Across words and participants, AnR stimuli resulted in the highest accuracy (58%), followed by AnS (37%) and An (24%). A linear mixed-effects model was constructed with dependent variable of phoneme accuracy, fixed effect of format, and participant and batch as random effects. There was a significant effect of format ($X^2_2 = 108$, $p < 10^{-16}$) driven by significant differences between all condition pairs (AnR *vs.* An, $t_{63} = -10.4$, $p = 10^{-11}$; AnS *vs.* An, $t_{63} = -4.5$, $p= 10^{-4}$; AnR *vs.* AnS, $t_{63} = -5.8$, $p = 10^{-6}$). These data show that both real and synthetic faces improve the intelligibility of noisy speech, with an advantage for real faces. Improvements in synthetic face animation could eventually reduce this advantage, allowing them to provide maximal benefit to listeners when human faces are unavailable. From a scientific perspective, synthetic faces are a valuable experimental tool because they allow different facets of talker identity, facial emotions, and speech mouth movements to be parametrically varied, manipulations that are difficult to achieve with real faces.

Disclosures: Y. Yu: None. **A. Lado:** None. **Y. Zhang:** None. **J. Magnotti:** None. **M.S. Beauchamp:** None.

Poster

PSTR152. Cross-Modal Processing in Humans

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR152.08/CC5

Topic: D.08. Multisensory Integration

Support: VBI Discovery Award
NSF DGE-1922697

Title: Contribution of stimulus parameter changes to auditory, visual, and audiovisual motion perception in humans

Authors: *A. TIESMAN[1], A. SCHOENHAUT[2], R. RAMACHANDRAN[3,2], M. T. WALLACE[1,2];
[1]Psychology, [2]Vanderbilt Brain Inst., Vanderbilt Univ., Nashville, TN; [3]Dept. of Hearing & Speech Sci., Vanderbilt Univ. Med. Ctr., Nashville, TN

Abstract: The ability to effectively integrate cues from multiple sensory modalities and combine them effectively into a unified percept of the world is vital to sensory processing; however, multisensory studies classically involve static stimuli, neglecting the fact that motion is prevalent in natural environments. The present study aims to investigate the impact of manipulating different physical attributes of auditory and visual motion stimuli on multisensory gain—the improvement in performance or the strengthening of neural responses when information from different senses is integrated compared to when each sense is considered independently. Using a two-alternative forced choice task, adult human participants were asked to report the direction of visual (random dot kinematogram), auditory (broad-band noise amplitude-modulated across two speakers and embedded in partially-correlated), and audiovisual stimuli. For audiovisual conditions, this task design intrinsically encourages participants to rely on both auditory and visual motion cues rather than attend to only one modality as in previous studies (Meyer & Wuerger, 2001; Kayser et al., 2017). To investigate the effects of stimulus properties, we first varied the motion strength (i.e., coherence) for the auditory and visual components when presented alone and together. We find that changing the motion coherence in turn creates a change in the degree of multisensory gain, which can be quantified by comparing responses on audiovisual trials to the responses of auditory and visual trials combined. Additionally, our results showed individual differences in how participants weigh auditory versus visual motion cues when both cues are available, which we quantified within the framework of maximum likelihood estimation (MLE). These individual differences could then be modeled to identify key factors that contribute to the participant's motion percept (e.g. prior trial direction, stimulus velocity, auditory dB SNR, etc.). Our findings lay the groundwork for future neurophysiological studies aimed at challenging what we know about multisensory motion processing in various contexts, such as attention.

Disclosures: A. Tiesman: None. **A. Schoenhaut:** None. **R. Ramachandran:** None. **M.T. Wallace:** None.

Poster

PSTR152. Cross-Modal Processing in Humans

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR152.09/CC6

Topic: D.08. Multisensory Integration

Support: NIH Grant DC016297

Title: Investigating how visual input helps solve a highly competitive cocktail-party problem

Authors: F. AHMED[1], ***A. NIDIFFER**[2], E. C. LALOR[2];
[1]Biomed. Engin., [2]Univ. of Rochester, Rochester, NY

Abstract: Real-life listening environments are often full of multiple competing speech sources, although listeners are typically interested in following just one of those. This is famously known as the 'cocktail-party problem'. Although there are numerous studies on cocktail-party problem, majority of these studies lack in one of three following ways- they 1) considered the cocktail-party problem in audio-only set up 2) are limited in the number of competing sound sources, restricted to only 1 distractor 3) used relatively simple stimuli such as syllables, isolated words, or short segments of speech. However, everyday speech communication consists of a dynamic flow of meaningful, connected words, along with various facial and articulatory gestures accompanying the ongoing acoustic information, and typically we are exposed to more than one distractor in real-world. Attending to desired speech therefore requires much more interaction between vision and audition that has been ignored in most existing studies. To explore how vision helps solve the cocktail party in more naturalistic scenarios, we have designed a paradigm where participants are presented with natural, continuous speech in a cocktail-party like scenario involving 5 simultaneous talkers while we record their EEG. The target talker appeared in both audio-only and audiovisual modality. We found that participants' ability to attend the target talker significantly improved when a congruent face accompanied the speech, and the improvement was also reflected in the cortical tracking of speech envelope. Further analyses will shed more light into the neural dynamics by which visual input facilitates our attention to desired speech.

Disclosures: F. Ahmed: None. **A. Nidiffer:** None. **E.C. Lalor:** None.

Poster

PSTR152. Cross-Modal Processing in Humans

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR152.10/CC7

Topic: D.08. Multisensory Integration

Title: Altered white matter morphology in age-related hearing loss

Authors: *S. ROSEMANN, C. M. THIEL;
Univ. of Oldenburg, Oldenburg, Germany

Abstract: Age-related hearing loss affects a large part of the older population and commonly affects the higher frequencies. Research using magnetic resonance imaging (MRI) provided evidence for neuroanatomical changes covering grey and white matter in age-related hearing loss. However, studies using diffusion-weighted magnetic resonance imaging (DWI) and a diffusion tensor imaging (DTI) model to compute measures such as fractional anisotropy (FA) or mean diffusivity (MD) showed inconsistent results concerning differences between normal-hearing and hard of hearing participants. Further, measures of FA do not account for the crossing fibers problem (multiple fiber populations within one voxel). Recently, more advanced diffusion models have been developed that can resolve these multiple fibers within a voxel. Hence, the aim of the current study was to investigate changes in white matter morphology in age-related hearing loss by employing a fixel-based approach to study the number of axons and the axon diameter within a voxel. Data from 28 hard of hearing and 31 normal-hearing participants (aged 50-75 years) were included in the analysis. The hard of hearing participants showed a uniformly varying degree of mild to moderate and symmetrical age-related hearing loss. DWI data were acquired with a multi-directional diffusion weighting (MDDW) sequence with 10 non-diffusion images and 45 diffusion images with a b-factor of b=3000 s/mm^2. We observed a significant decrease in fiber density (FD, indicator for a change in tissue microstructure) for fiber bundles originating in the body of the corpus callosum (fronto-medial fibers) and in the cerebellum in hard of hearing compared to normal-hearing participants. Further, we found a significant decrease in the combined measure of fiber density and fiber bundle cross section (FDC, indicating microscopic and macroscopic changes) in a small part of frontal projecting fibers from the corpus callosum in hard of hearing participants. Our data provide evidence of reduced FD and FDC in age-related hearing loss, possibly indicating a loss of axons as well as decreased axon diameter in fibers that originate in the corpus callosum and project to the medial part of the precentral gyrus. This is the first study assessing whiter matter morphology with an advanced diffusion model resolving fiber crossings. These results suggest that age-related hearing loss is associated with a loss of tissue microstructure in the main interhemispheric commissure.

Disclosures: S. Rosemann: None. **C.M. Thiel:** None.

Poster

PSTR152. Cross-Modal Processing in Humans

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR152.11/CC8

Topic: D.08. Multisensory Integration

Support: NIH NIDCD R01 DC020717

Title: Speech representation in Heschl's gyrus modified by visual input

Authors: *I. DEWITT[1], A. MAJUMDAR[2], C. CAO[2], W. C. STACEY[3], D. BRANG[2]; [2]Psychology, [3]Neurol., [1]Univ. of Michigan, Ann Arbor, MI

Abstract: Lipreading can augment and facilitate speech perception. The neural mechanisms of multimodal processing in speech perception remain poorly understood. To investigate the hypothesis that visual input sharpens speech sound representations in auditory cortex, we recorded intracranial EEG (iEEG) data from patients with intractable epilepsy during an audiovisual word identification task. In the task, participants heard and-or saw 16 spoken CVC words formed by combining 4 initial phonemes (/m, n, b, g/) with 4 trailing diphones (/ih-l, ey-t, ae-sh, uw-n/). To assess phoneme representations in auditory cortex, we trained SVM classifiers to decode phoneme identity for initial and medial phonemes (/m, n, b, g, ih, ey, ae, uw/) from event-related potentials (ERPs) recorded from contacts in Heschl's gyrus. Consistent with visual speech increasing the precision of neuronal representations in auditory cortex, audiovisual speech showed higher decoding accuracy compared to auditory-only speech. Time-windowed analyses showed that phoneme identity information was decodable from the auditory system immediately after phoneme onset for approximately 800 ms, with initial and medial phonemes decodable from a large overlapping window of time-points. Moreover, we found enhanced decoding ability for audiovisual stimuli (relative to audio-alone stimuli) shortly after phoneme onset and lasting up to 300-ms post phoneme onset. Preliminary analyses of band-limited spectral-channels (including high-gamma power) did not indicate ERP decoding performance was driven by power in a single spectral channel, suggesting oscillatory phase may contribute to ERP decoding performance. Classification accuracy time course and the spatial distribution of contributing electrodes imply that initial and medial phoneme representations are simultaneously active.

Disclosures: I. DeWitt: None. **A. Majumdar:** None. **C. Cao:** None. **W.C. Stacey:** None. **D. Brang:** None.

Poster

PSTR152. Cross-Modal Processing in Humans

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR152.12/CC9

Topic: D.08. Multisensory Integration

Support: ERC Consolidator Grant (PAINSTRAT)

Title: The Vertex Potential exert a phase-dependent modulation of both sensory and motor brain function

Authors: *S. PEROVIC[1,2], R. SOMERVAIL[1], G. IANNETTI[1,3];
[1]Italian Inst. of Technol., Rome, Italy; [2]Dept. of Physiol. and Pharmacol., Sapienza Univ. of Rome, Rome, Italy; [3]Dept. of Neuroscience, Physiol. and Pharmacol., Univ. Col. London, London, United Kingdom

Abstract: Sudden and isolated changes in the sensory environment elicit a transient, large, and widespread negative-positive deflection in the ongoing electroencephalogram: the Vertex

Potential (VP). In the past decades VP has been interpreted as a result of modality-specific sensory processing. However, current research showed that this response is supramodal, encodes the stimulus novelty and behavioural relevance, and is tightly related to modulation of ongoing motor activity. For this reason, we hypothesize that VP is involved in regulating global arousal and modulating ongoing brain activity. If the VP truly reflects a global modulation of brain activity, it should not only influence the motor system, but its effects should be more pervasive. To test this hypothesis, we investigated the influence of VP on somatosensory processing: we recorded early-latency somatosensory evoked potentials (SEPs) elicited by continuous high-frequency median nerve stimulation while VPs were concomitantly elicited by isolated auditory stimuli in 23 healthy human participants. The amplitude of the N20 SEP component, which represents the first arrival of the thalamocortical volley to the primary somatosensory cortex, was unaffected by the VP. In contrast, the amplitude of the P45 component, which reflects later cortical processing, was reduced during the negative and enhanced during the positive VP wave. These results provide strong evidence that the VP exerts phase-dependent modulation of early sensory processing. The biphasic nature of this modulation is remarkably similar to the VP-dependent modulation of corticospinal output (fig.1), indicating that its effect is not limited to a single brain system, but it is rather a global phenomenon.

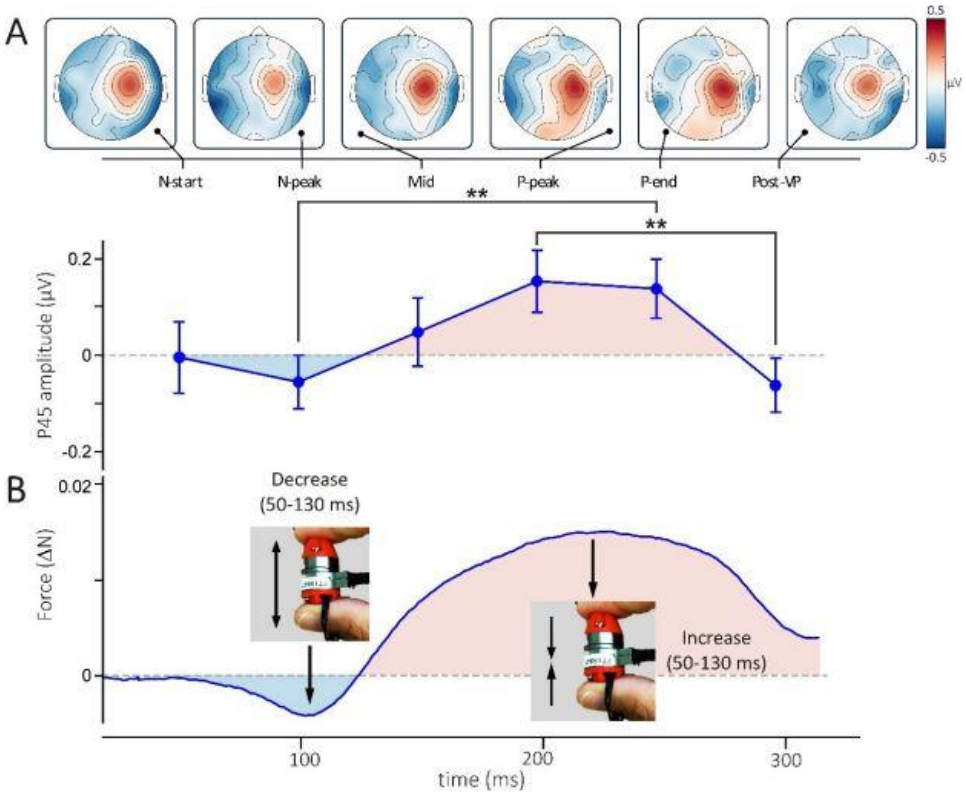


Figure 1. A. VP phase-dependent modulation of P45 SEP component: the P45 amplitude shows a biphasic modulation, dependent on the when it occurred during the VP phase. There was strong evidence of a difference between P45 amplitude during the negative compared to the positive wave of the VP. The top panel shows the scalp distribution of the P45 wave at different VP phases. Note that the P45 component is contralateral to the stimulated hand, and is smaller during the negative and larger during the positive wave of the VP. **B.** The VP exerts a remarkably similar phase-dependent modulation of isometric force, which decreases during the negative and increase during the positive VP wave (adapted from Novembre et al. *J Neuroscience* 2018).

Disclosures: **S. Perovic:** None. **R. Somervail:** None. **G. Iannetti:** None.

Poster

PSTR152. Cross-Modal Processing in Humans

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR152.13/CC10

Topic: D.08. Multisensory Integration

Support: R01 DC-013543

Title: Neural Mechanisms of the Audiovisual Time-Flow Illusion

Authors: *M. GONZALES, K. C. BACKER, A. J. SHAHIN;
Univ. of California, Merced, Merced, CA

Abstract: The Audiovisual Time-Flow Illusion (ATFI) is an auditory-induced visual illusion whereby "pausing" or "skipping" in the visual stream is perceived when temporally intact visual speech is merged with pausing or skipping auditory speech, respectively. In our behavioral account of this phenomenon (Gonzales et al., 2022), results showed that visual "pausing" was perceived 35.4% of the time when pausing audio was combined with an unchanged video. Similarly, visual "skipping" was perceived 48% of the time when skipping audio was combined with an unchanged video. Both illusory percepts were experienced significantly above chance level. When stimulus conditions were reversed and temporally intact audios were combined with pausing or skipping videos, no visually-induced auditory illusion was observed. The present study aims to investigate the neural mechanisms underlying the ATFI, using EEG. Preliminary EEG results revealed significant alpha wave suppression and increased theta wave activity over occipital and fronto-central regions directly following the onset of the auditory abruptions (onset of auditory skipping or pausing). Notably, occipital electrodes were found to have greater alpha suppression and a greater increase of theta activity. As of now, we cannot determine if this neural activity is auditory and/or visual in nature, pending further analysis to determine the modality. Nonetheless, these results suggest that there is a distributed network of activity reflected in theta and alpha dynamics that drives the ATFI, such that the visual system is reengaged as visual information is realigned relative to the auditory stream. This is in accordance with natural speech processing in which mouth movements typically precede sound production; consequently, the visual modality has a predictive (leading) role in processing unfolding speech streams. Thus, the ATFI reflects the visual modality's attempt to realign itself in order to assume a leading position relative to the auditory stream.

Disclosures: M. Gonzales: None. **K.C. Backer:** None. **A.J. Shahin:** None.

Poster

PSTR152. Cross-Modal Processing in Humans

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR152.14/CC11

Topic: D.08. Multisensory Integration

Support: ISF-NSFC Grant 3318/20

Title: Neural correlates of heading ambiguity

Authors: *R. KARNI, E. ZION GOLUMBIC, A. ZAIDEL;
Gonda Multidisciplinary Brain Res. Ctr., Bar-Ilan Univ., Ramat Gan, Israel

Abstract: Perceptual decision-making requires accumulating sensory evidence. However, sensory information is often ambiguous and noisy. Hence, the brain must disambiguate sensory input and extract decision-relevant information. Here, we used EEG to explore the neural correlates of perceptual ambiguity in perceiving one's own motion in space (self-motion perception). Participants performed a two-alternative forced-choice (2AFC) task of heading discrimination, in which they were required to report whether a self-motion stimulus was rightward or leftward of straight-ahead. Stimuli were generated using a motion simulator and comprised one of two sensory cues: vestibular (inertial motion of the 3D motion platform) or visual (optic flow, via a head-mounted virtual reality display). The heading stimuli were either: 1) relatively easy to discriminate ('unambiguous'; ±8° relative to straight ahead) or 2) hard to discriminate. ('ambiguous'; 0° forward motions straight ahead). Participants were always required to choose whether their self-motion was rightward or leftward, even for the ambiguous stimulus. We analyzed event-related potentials (ERPs) and event-related spectral power (ERSP) of the centro-parietal area for ambiguous vs. unambiguous stimuli for both cues (visual and vestibular). The ERP signals increased slower, peaking about 200ms later for ambiguous vs. unambiguous stimuli. The ERSP induced greater and longer alpha event-related desynchronization (ERD) for the ambiguous vs. unambiguous signals in the 500-1000ms post-stimulus onset. These results expose neural correlates of perceptual ambiguity in centro-parietal EEG signals. Ambiguous stimuli were accompanied by delays in ERP and extended ERD. This observation is in line with delayed/extended processing of stimuli, presumably required to disambiguate the ambiguous stimuli. Moreover, similar differences between responses to ambiguous vs. unambiguous stimuli were seen for both cues (vestibular and visual). This suggests a general (supramodal) signature of perceptual ambiguity. This neural signature of ambiguity can help detect ambiguous states in real-life decisions, such as driving, and can be useful for developing assistive brain-computer interfaces (BCI).

Disclosures: **R. Karni:** None. **E. Zion Golumbic:** None. **A. Zaidel:** None.

Poster

PSTR152. Cross-Modal Processing in Humans

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR152.15/CC12

Topic: D.08. Multisensory Integration

Support: NIH Grant R01NS102920-02
Dana & Albert R. Broccoli Charitable Foundation
Nanette and Burt Forester
Roberta Wilson

Title: Progressive reduction of exoskeleton robotic assistance in stepping after a complete spinal injury when facilitated with transcutaneous spinal neuromodulation and training

Authors: *I. MONTOYA[1], H. R. TORRES SOLANO[2], K. CHANG[2], H. ZHONG[2], K. KIJIMA[2], V. EDGERTON[2];
[1]California State Univ. Los Angeles, MARINA DEL REY, CA; [2]Rancho Res. Inst., Downey, CA

Abstract: The primary objective of this case study is to determine whether an individual with a complete spinal cord injury can regain independent stepping ability by using an exoskeleton (Ekso Bionics NR), electrical spinal neuromodulation, and progressive training of multiple sensory-motor skills to reestablish functional supraspinal-propriospinal-spinal bidirectional connectivity. Experiments from multiple laboratories using a noninvasive, transcutaneous strategy to neuromodulate the spinal cord have consistently demonstrated that individuals diagnosed clinically as having a complete spinal cord injury can regain functionally novel connectivity that enables voluntary motor control. This includes stepping, improved trunk control, coughing, and upper limb dexterity. Additional improvements in organ systems predominantly under autonomic control have been shown as well. The subject of this case study is 10 years post-injury and suffers from a unilateral brachial plexus nerve root avulsion injury and a thoracic spinal cord injury at the T4/T5 level. The subject has used this combination of neuromodulation and training consistently over a 2-year period, 2-3x/week for approximately 45 min/intervention. Non-invasive spinal stimulation techniques have enabled partial restoration of stepping while assisted on a treadmill. The exoskeleton has been programmed to adaptive settings to allow for the subject to learn to control components of the step cycle of each lower limb with increasing levels of control. This design allows for the quantification of the amount of work that is performed by the subject in relation to the device. To date the subject has demonstrated up to a 55% increase in the relative work provided by both limbs to complete a step in the exoskeleton. In addition, robust levels of EMG bursts have emerged in flexor and extensor muscles of the proximal (hamstrings, rectus femoris) and distal (tibialis anterior and soleus) limb segments. These patterns of EMG activity reflect a more normal agonist-antagonist reciprocal pattern typical of flexor and extensor motor pools. Equally important, the emergence of persistent sensations has enabled the subject to detect temporally accurate placement of each foot during stepping. These findings have the potential to enhance the development of personalized and effective neurorehabilitation strategies for individuals with severe spinal cord injuries, thereby improving their quality of life and functional independence.

Disclosures: I. Montoya: None. **H.R. Torres solano:** None. **K. Chang:** None. **H. Zhong:** None. **K. Kijima:** None. **V. Edgerton:** E. Ownership Interest (stock, stock options, royalty, receipt of intellectual property rights/patent holder, excluding diversified mutual funds); Part ownership or ONWARD and SpineX Inc.

Poster

PSTR152. Cross-Modal Processing in Humans

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR152.16/CC13

Topic: D.08. Multisensory Integration

Support: VERTICES NIH Grant 5R25GM134979-03
VBI Training Grant 5U54HD083211

Title: The influence of audiovisual stimuli on sound localization in individuals with temporary unilaterally attenuated hearing

Authors: *M. K. STEVENS[1], S. G. VASSALL[1], M. T. WALLACE[2];
[2]Hearing and Speech Sci., [1]Vanderbilt Univ., Nashville, TN

Abstract: Multisensory integration—the process by which information from the different sensory modalities is combined by the brain—not only creates our perceptual gestalt, but also results in profound behavioral benefits in the natural environment. However, when sensory cues are incongruent, they can often result in illusory percepts, such as in the case of the ventriloquism effect. The ventriloquism effect occurs when auditory and visual cues are temporally synchronous but spatially incongruent. In this effect, when asked to localize the source of an auditory stimulus, most individuals show a bias toward the visual stimulus' location. Importantly, auditory localization depends on input from both ears, and if hearing in one ear is compromised—as in the case of unilateral hearing loss—auditory localization accuracy decreases. However, it remains unknown how the presence of a simultaneous visual stimulus affects auditory localization in this population. To begin to answer this question, five normal-hearing participants performed an auditory localization task in which they were asked to report the location of an auditory white noise burst presented either alone or accompanied by a spatially congruent or incongruent visual stimuli (i.e., flash). Participants completed this task both normally and with one ear plugged to simulate unilateral hearing attenuation. Although preliminary data shows no group-level performance differences in auditory-only or audiovisual performance across conditions, some participants did show significantly decreased performance on auditory-only accuracy with one ear plugged. These same subjects showed a significantly increased bias toward the visual stimulus in the incongruent audiovisual condition. Additionally, despite a high level of heterogeneity across participants, in line with prior literature, all individuals showed an increased visual bias at smaller audiovisual disparities, with a gradual decrease in bias as distance between the auditory and visual cues increased. This preliminary data suggests that some individuals are highly sensitive to unilateral hearing attenuation, and that when spatial hearing is disrupted, the visual stimulus becomes more important for informing the perception of the auditory cue location. In the future, we aim to increase the sample size to gain a better understanding of the range of individual auditory spatial hearing abilities, as well as recruit a true clinical population of individuals with unilateral hearing loss.

Disclosures: M.K. Stevens: None. **S.G. Vassall:** None. **M.T. Wallace:** None.

Poster

PSTR152. Cross-Modal Processing in Humans

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR152.17/CC14

Topic: D.08. Multisensory Integration

Support: R01NS127777

Title: Population-level coding of multisensory frequency signals in human neocortex

Authors: A. MACKLIN[1], K. PERKS[2], L. WANG[3], M. O'MALLEY[1], *J. YAU[3];
[1]Rice Univ., Houston, TX; [2]Univ. of Washington, Seattle, WA; [3]Baylor Col. of Med., Houston, TX

Abstract: Humans perceive the frequency of environmental oscillations by audition (sound waves) and touch (mechanical vibrations). Audition and touch are known to interact in temporal frequency perception. Auditory and tactile frequency signals may be represented in the same brain regions or by the same neural populations. Despite the perceptual interactions, it remains unknown what operations are performed by neural populations to combine auditory and tactile information. Here, we determined how unisensory and multisensory frequency signals are distributed over cortical sensory hierarchies and what computations support multisensory processing. We used functional magnetic resonance imaging (fMRI) and measured fMRI BOLD responses in human participants experiencing vibrations only, sounds only, and audio-tactile stimulus combinations. First, we implemented a hierarchical clustering analysis to characterize unisensory and multisensory response profiles. Initial results reveal that distinct response motifs are distributed across auditory, somatosensory and premotor cortices in a manner consistent with hierarchical processing. Second, we implemented competing encoding models to predict voxel-level responses to the multisensory cues. Preliminary modeling results reveal that multisensory responses are most consistent with cue integration in frontal and parietal regions and with conjunction coding in temporal regions. These findings establish a computational framework for understanding multisensory frequency signals and their distribution across the human brain.

Disclosures: A. Macklin: None. **K. Perks:** None. **L. Wang:** None. **M. O'Malley:** None. **J. Yau:** None.

Poster

PSTR152. Cross-Modal Processing in Humans

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR152.18/CC15

Topic: D.08. Multisensory Integration

Support: NSERC Discovery Grant

Title: Optimizing a temporal range for visual-tactile crossmodal enhancement in early cortical somatosensory processing

Authors: *E. SALAZAR[1], M. V. FAERMAN[2], M. S. DAUB[2], S. THOMPSON[2], S. K. MEEHAN[3], R. STAINES[4];

[1]Kinesiology and Hlth. Sci., [3]Dept. of Kinesiology and Hlth. Sci., [2]Univ. of Waterloo, Waterloo, ON, Canada; [4]Univ. Waterloo, Waterloo, ON, Canada

Abstract: Bimodal interactions between relevant visual and tactile inputs can facilitate attentional modulation at early stages in somatosensory cortices to achieve goal-oriented behaviours. However, the specific neural mechanisms contributing to this modulation are unclear. We used electroencephalography (EEG) to observe the temporal contributions of visual priming to the enhancement of somatosensory cortical responses. We hypothesized that somatosensory activity would be modulated based on the temporal onset of visual stimuli in task-relevant bimodal (visual-tactile) events and specifically would be enhanced by visual inputs with an onset of at least 200 ms prior to the tactile event. Somatosensory modulation was inferred through amplitude and latency shifts in tactile event-related potentials (ERPs) recorded while participants performed a sensory integration task that required scaled motor responses dependent on the amplitudes of tactile and visual stimuli. Tactile stimuli were discrete vibrations (25 Hz) presented to the index finger, visual stimuli were presented as a central horizontal bar on a computer screen at varying heights, and graded motor responses were made by squeezing a pressure-sensitive rubber bulb. Healthy adults completed a training session to become familiar with the stimulus-response relationships for both visual and tactile stimuli and 1 of 3 experiments that presented pairs of discrete stimuli with random amplitude variations: tactile-tactile (TT; 500 ms each, 30 ms ISI), visual-tactile with simultaneous onset (SIM), and visual-tactile with a delayed latency (VTd; 500 ms each). In the 3 experiments (n=34) participants were presented with various random delays between visual-tactile stimulus onsets for the VTd condition (0-100 ms, 100-200 ms, 200-300 ms, 300-400 ms). Two of these studies measured ERPs from the right somatosensory cortex (S1)(left index, right hand on bulb), while the other measured ERPs from the left S1 (right index, left hand on bulb). In all cases the experimental task was to summate the 2 stimuli presented per trial and represent the intensity by a graded response. In the last study, an additional task where only the visual component is responded to was also done to further address the effects of attentional modulation. The 200-300 ms temporal window showed the largest and most consistent upregulation of early cortical somatosensory processing, represented by an increase in P50 amplitude ($p<0.01$). These results enhance our understanding of the S1 temporal processing and establish a foundation to explore the behavioural effects and interactions with the attentional network associated with such S1 enhancements.

Disclosures: E. Salazar: None. **M.V. Faerman:** None. **M.S. Daub:** None. **S. Thompson:** None. **S.K. Meehan:** None. **R. Staines:** None.

Poster

PSTR152. Cross-Modal Processing in Humans

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR152.19/CC16

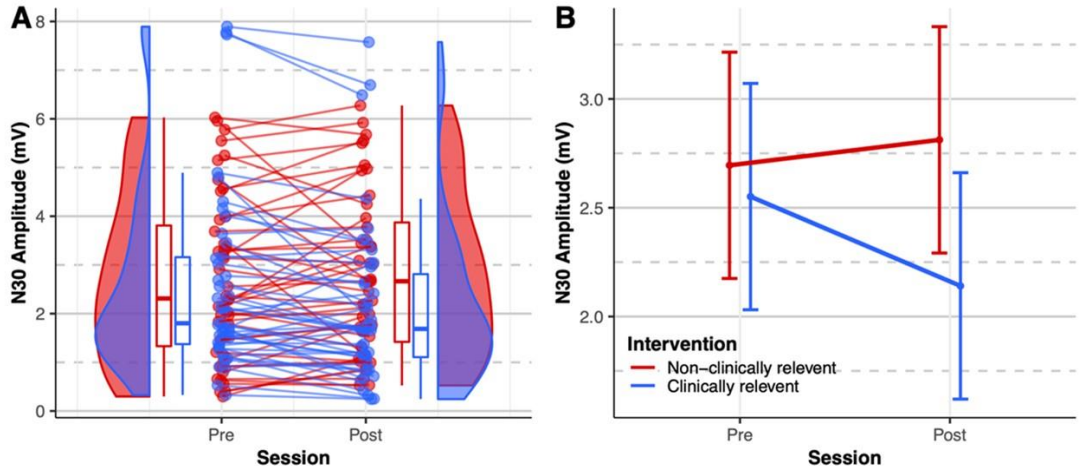
Topic: D.08. Multisensory Integration

Support: Australian Spinal Research Foundation

Title: Directing a specific adjustive thrust towards a chiropractic subluxation significantly alters sensorimotor integration compared to directing a thrust at a non-subluxated vertebrae

Authors: *H. HAAVIK[1], K. HOLT[2], N. KUMARI[3], C. MERKLE[3], I. AMJAD[3], S. NAVID[3], I. K. NIAZI[2];
[1]NewZealand CollegeChiropractic, Auckland, New Zealand; [2]Ctr. for Chiropractic Res., [3]New Zealand Col. of Chiropractic, Auckland, New Zealand

Abstract: Objective: This study aimed to compare neurophysiological outcomes of an adjustive thrust directed at a chiropractic subluxation in the upper cervical spine compared to a thrust directed at a upper cervical motion segment that did not display any clinical indicators of dysfunction. **Methods:** In this parallel group randomized controlled trial, 86 participants with evidence of a chiropractic subluxation in their upper cervical spine (C1-3), were randomly allocated to receive a specific adjustive thrust (using an Activator Adjusting Instrument) to a subluxated upper cervical vertebrae or to an upper cervical vertebrae that did not display any clinical indicators of dysfunction. A motion segment was deemed to be subluxated when the following clinical indicators were present: tenderness to palpation, restricted intersegmental motion, asymmetric muscle tension, and blocked joint-play or end-feel.. Somatosensory evoked potentials (SEPs) from median nerve stimulation were recorded before and immediately after the intervention. The amplitude of the N30 SEP peak (an indicator of early sensorimotor integration; SMI) was the primary outcome measure. **Results:** A linear mixed-model revealed a significant interactive effect between site of intervention and session ($F(1,84)=9.89$, $p<0.002$). Pairwise comparisons using Tukey's HSD showed that there was a statistically significant N30 peak amplitude decrease (by 17%, p=0.005) in the subluxation adjustment group (−16.76±28.32%). There was a non-significant increase in the N30 SEP amplitude in the non-subluxation thrust group (19.58%±55.09%, p=0.757). **Conclusion:** This study showed that directing a specific adjustive thrust to a chiropractic subluxation resulted in significant neurophysiological changes in SMI similar to those observed in previously published papers, while thrusts directed at non-subluxated segments had no significant effect.



Disclosures: **H. Haavik:** None. **K. Holt:** None. **N. Kumari:** None. **C. Merkle:** None. **I. Amjad:** None. **S. Navid:** None. **I.K. Niazi:** None.

Poster

PSTR152. Cross-Modal Processing in Humans

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR152.20/CC17

Topic: D.08. Multisensory Integration

Support: Dana and Albert R. Broccoli Charitable Foundation
Nanette and Burt Forester

Title: Noninvasive spinal neuromodulation and subject specific rehabilitation of child with cerebral palsy (CP) triggers multiple system neural networks that reorganizes and enhances multiple sensory-motor behaviors

Authors: *S. HASTINGS[1], K. CHANG[2], H. ZHONG[2], J. GONNELLA[1], C. GONNELLA[1], V. EDGERTON[2];
[1]Susan Hastings Pediatric Physical Therapy, San Jose, CA; [2]Rancho Res. Inst., Downey, CA

Abstract: We hypothesized that a combination of noninvasive spinal electrical neuromodulation and a subject specific activity-training program can engage proprioception input that can transform dysfunctional networks of a child diagnosed as CP spastic diplegia to highly, relatively normal functional states. The child had previously used common physical therapy treatments focused on control of the center of mass. A targeted treatment began at age of 19 months, where his Gross Motor Functional Classification Scale (GMFCS) improved from Level 2 to Level 1. He did not undergo any recommended surgeries or pharmacological interventions. At age 11, a noninvasive Transcutaneous Spinal Neuromodulation (TSN) strategy was initiated in combination with play activities consistent with his age and interests.
Results:
Multiple, complex motor skills were performed with a gravitationally aligned posture, such as stepping 5.25 km in 65 min (4.7km/hr.), sprinting at 9.7kh/hr for 1 min without undue fatigue, stepping backwards and sideways, jumping rope and whirling a Hoola-hoop while stepping on a treadmill and jumping 56 cm high. Clarity and speaking speed , improved swallowing and hand writing also became evident immediately with neuromodulation. The Selective Control of the Lower Extremities and 6-Minute Walk Test scores reached population limits for typically developing children. After a range of standard rehabilitative treatments up to age 11, new and complex skills, previously attempted without success, emerged with the introduction of spinal neuromodulation combined with rehabilitation.
Interpretation:
While children with CP generally can perform most movements, they are markedly dysfunctional, stereotypical, and commonly subject to spasticity, reflecting a functionally abnormal spinal-supraspinal connectivity. While it is generally assumed that most of these

dysfunctions can be attributed primarily to supraspinal networks, we propose that the more normal connectivity that persist between peripheral proprioception-cutaneous input to the spinal networks can be used to guide the reorganization of a more normal spinal- supraspinal connectivity. The level of plasticity necessary to achieve the required reorganization of within and among different neural networks, was achieved with a combination of TNS and specific activity-dependent mechanisms. By engaging these two concepts we have demonstrated in a single patient a strategy that can lead to a successful adaptation of bidirectional reorganization of proprioception-spinal cord-brain connectivity to higher levels of functionality, without any invasive surgical or pharmacological interventions.

Disclosures: **S. Hastings:** None. **K. Chang:** None. **H. Zhong:** None. **J. Gonnella:** None. **C. Gonnella:** None. **V. Edgerton:** E. Ownership Interest (stock, stock options, royalty, receipt of intellectual property rights/patent holder, excluding diversified mutual funds); Part ownership in ONWARD and SpineX.

Poster

PSTR152. Cross-Modal Processing in Humans

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR152.21/CC18

Topic: D.08. Multisensory Integration

Support: NIH R01NS065395
NIH U01NS113339

Title: Multivariate Neural Measures of Speech Intelligibility in fMRI and iEEG

Authors: *J. MAGNOTTI[1], Y. ZHANG[1,2], X. ZHANG[1], A. A. SANGANI[1], Z. WANG[1], M. S. BEAUCHAMP[1];
[1]Univ. of Pennsylvania, Philadelphia, PA; [2]Baylor Col. of Med., Houston, TX

Abstract: Humans have the unique ability to decode the rapid stream of language elements that constitute speech. Although auditory noise in the environment interferes with speech perception, visual speech from the face of the talker may compensate. However, individuals vary in their use of visual speech and the amount of visual information varies based on the speech content. We combined behavior, BOLD fMRI, and intracranial EEG to examine the neural correlates of these person-level and word-level differences. Fifty-two participants were presented with speech in four formats: audiovisual speech (AV) with/without auditory noise and auditory-only (AO) speech with/without noise. In line with past research, clear speech was highly intelligible, noisy speech was sometimes intelligible, and AV speech was more intelligible than AO speech. Trials were sorted based on intelligibility, and multivariate analyses were used to compare the patterns of activity in superior temporal cortex evoked by clear speech (without added auditory noise) and intelligible vs. unintelligible noisy speech. For BOLD fMRI participants (n = 37), we used all voxels in superior temporal cortex in each hemisphere. For iEEG patients (n = 15), we found 140

temporal electrodes that showed a significant response to clear AO speech (mean 70-150Hz broadband high frequency activity from 0-1s following auditory onset vs. baseline, $p < 0.001$ Bonferroni corrected). For both BOLD fMRI and iEEG, pairwise pattern correlations between clear speech and intelligible noisy speech were higher than the correlation between clear speech and unintelligible noisy speech. Using the larger sample size of the fMRI data, we found that differences in multivariate pattern similarity (as indexed by multidimensional scaling) corresponded to intelligibility of noisy speech for both words and sentences, for both AO and AV speech ($r = 0.55$). Using the greater temporal resolution of the iEEG data, we found that word-level differences in intelligibility were predicted by neural pattern similarity: when the response to a noisy word was more similar to the response to the clear version of that word, perceptual intelligibility was higher ($r = 0.43$). Additionally, we found that the neural pattern improvement for AV vs. AO speech reliably predicted the degree of audiovisual improvement for that word, presumably as a result of the neural integration of the viseme and phoneme content of each word. Understanding the neural substrates of individual- and word-level differences in noisy speech perception may help to develop strategies for helping those with impaired speech perception.

Disclosures: **J. Magnotti:** None. **Y. Zhang:** None. **X. Zhang:** None. **A.A. Sangani:** None. **Z. Wang:** None. **M.S. Beauchamp:** None.

Poster

PSTR152. Cross-Modal Processing in Humans

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR152.22/CC19

Topic: D.08. Multisensory Integration

Support: European Union's Horizon 2020 Research and Innovation Program under Marie Skłodowska-Curie Actions ITN Grant Agreement No. 860114 (Project multiTOUCH)
the Belgian Excellence of Science (EOS) program (Project No. 30991544)

Title: Aligned motion-direction information for touch and vision in hMT+/V5

Authors: *I. SHAHZAD[1], C. BATTAL[1,3], F. CERPELLONI[2,4], A. VAN AUDENHAEGE[1], A. MOURAUX[2], O. COLLIGNON[1,2,5];
[1]Inst. of Neurosci. (IoNS), [2]Inst. of Res. in Psychological Sci. (IPSY), Univ. Catholique de Louvain, Brussels, Belgium; [3]Univ. Catholique de Louvain, Inst. of Res. in Psychological Sci. (IPSY), Brussels, Belgium; [4]Katholieke Univ. Leuven, Leuven, Belgium; [5]HES-SO Valais-Wallis, The Sense Innovation and Res. Ctr., Brussels, Belgium

Abstract: Motion directions can be perceived through vision and touch. This motion information coming from the different senses must spatially align to carry out optimal goal-directed tasks. The alignment is not trivial since vision and touch are initially coded in different spatial frames

of references and the body parts can change postures, which eventually change the external coordinates while keeping the somatotopic coordinates intact. Is there any brain region where the direction of visual and tactile motion is coded using a common frame of reference independent of body-posture? To address this unsolved question, we characterized the brain activity of participants using fMRI in two experiments. We first implemented visual and tactile motion localizers to functionally identify motion selective regions in each individual in both the modalities. In addition to sensory specific motion selective regions, whole-brain univariate analyses revealed that the middle occipito-temporal region (hMT+/V5) is motion selective across the senses. In another experiment, we delivered directional visual and tactile motion stimuli across different hand postures. Multivariate pattern analyses revealed that motion directions can be decoded in both vision and touch in hMT+/V5. Importantly, information about tactile directions was enhanced when mapped using an externally-centred coordinate system as compared to a somatotopic frame of reference in this region. Finally, crossmodal decoding showed that tactile directions defined using an externally-centred coordinate system, but not the somatotopic frame of reference, align with the representation of visual directions in hMT+/V5. Our results show that motion directions in vision and touch are aligned in hMT+/V5 relying on a common external frame of reference. hMT+/V5, as a motion-selective region across different sensory modalities, may play a role in transforming spatial frames of reference during multisensory motion processing.

Disclosures: I. Shahzad: None. **C. Battal:** None. **F. Cerpelloni:** None. **A. Van Audenhaege:** None. **A. Mouraux:** None. **O. Collignon:** None.

Poster

PSTR152. Cross-Modal Processing in Humans

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR152.23/CC20

Topic: D.08. Multisensory Integration

Support: CRSNG
Chaire Fondation Caroline Durand en audition et vieillissement de l'Université de Montréal

Title: Dancers modulate the weighting of different sensory modalities to maintain postural control under challenging condition

Authors: *D. PAROMOV[1,2], K. MOÏN-DARBARI[1,2,3], M. MAHEU[1], B.-A. BACON[4], F. CHAMPOUX[1,2];
[1]Univ. de Montréal, Montréal, QC, Canada; [2]Ctr. de recherche de l'Institut Universitaire de Gériatrie de Montréal, Montréal, QC, Canada; [3]Montreal Ctr. for Interdisciplinary Res. in Rehabil., Montréal, QC, Canada; [4]Carleton Univ., Ottawa, ON, Canada

Abstract: Postural control is a complex motor skill that requires the integration of visual, vestibular, and somatosensory information to maintain balance. Previous studies have suggested differences in sensory strategies between professional dancers and non-dancers. Although some studies show that dance training allows for more rapid adaptation to vestibular stimulation, the effects of this type of stimulation remain poorly explored. The aim of the present study was to assess the effect of long-term multisensory training (i.e. dance training) on sensory reliance during passive vestibular stimulation. Twenty-four subjects with normal auditory and vestibular function were asked to perform a static postural control task under four conditions (Eyes open/closed; standing barefoot on a firm/foam surface) while galvanic vestibular stimulation (GVS) was induced. The analysis confirms significant differences in the weighting given to each of the senses between dancers and non-dancers as they strive to maintain postural control during vestibular perturbations. The data also provide more insights about the processes leading to the improved performance of dancers in challenging postural control tasks.

Disclosures: D. Paromov: None. **K. Moïn-Darbari:** None. **M. Maheu:** None. **B. Bacon:** None. **F. Champoux:** None.

Poster

PSTR152. Cross-Modal Processing in Humans

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR152.24/DD1

Topic: D.08. Multisensory Integration

Support: DFG Project ID 122679504 – SFB 874

Title: Phantom touch illusion: Unexpected phenomenological effects of visual touch in the absence of tactile input

Authors: *A. PILACINSKI, M. METZLER, C. KLAES;
Ruhr-University Bochum, Bochum, Germany

Abstract: You can’t tickle yourself. If you try sliding a finger along your forearm, the tickle sensation will be much weaker than if there was an insect crawling down your skin. This is because the nervous system attenuates the predicted sensory input caused by your own movements (Blakemore et al., 2000). This mechanism is called tactile gating/tactile gating. But what happens with tactile attenuation if there is no afferent tactile signal? Here, we tested this using an immersive virtual reality (VR) scenario in which subjects touched their body using a virtual object. This touch resulted in a tingling sensation corresponding to the location touched on the virtual body. We called it phantom touch illusion (PTI). The subjectively-reported intensity of the illusion has different strength across different parts of the hand. Interestingly, the illusion was also present when subjects touched invisible (inferred) parts of their limb. We reason that PTI results from the tactile gating process during self-touch. We additionally tested this hypothesis while comparing PTI to self-touch using different laser pointers and pantomimed

(no visible effector) touch. These conditions were reported as touch sensation by a significantly lower proportion of subjects. Importantly, subjects did not report tingling in these laser stimulation conditions, but rather other sensations, such as thermal, consistent with prior literature e.g. on “butcher tongue illusion”. The presence of PTI when touching invisible body parts suggests that tactile attenuation might be not exclusively based on vision, but rather on multi-sensory input involving body schema. Lastly, the presence of different phenomenal qualities during self-touch using a laser pointer might indicate top-down influences on phantom touch through cognitive mechanisms such as phenomenological control or sensory suggestibility. It remains to be determined, to what degree PTI is controlled by such cognitive mechanisms.

Disclosures: **A. Pilacinski:** None. **M. Metzler:** None. **C. Klaes:** None.

Poster

PSTR152. Cross-Modal Processing in Humans

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR152.25/DD2

Topic: D.08. Multisensory Integration

Support: UMass Boston Undergraduate Research Funds

Title: Attention influences neural responses based on crossmodal correspondences: Responses to abstract shapes are enhanced even when attending irrelevant shape features depending on audio-visual congruency

Authors: *E. MORINA, M. BESHARA, V. M. CIARAMITARO;
Dept. of Psychology, Univ. of Massachusetts, Boston, Boston, MA

Abstract: Crossmodal correspondences refer to the systematic mappings of features between different sensory modalities (e.g., vision, audition, touch, and smell). One example is the bouba-kiki effect, where abstract shapes are associated with nonsense words, specifically round shapes with /bouba/ sounds and spiky shapes with /kiki/ sounds. We used EEG frequency tagging, with fast periodic visual stimulation, to quantify how neural responses to a given visual shape depend on whether the shape is attended vs unattended or how auditory features match (congruent) or do not match (incongruent) visual features. We hypothesized neural responses would be enhanced when a shape was attended vs unattended, and when a sound was congruent (eg, /ki/) vs incongruent (eg, /ba/) to the attended shape (eg, spiky). Sixteen participants (18-35-year-old; 9 females) viewed a shape (round or spiky), frequency modulated in brightness (5.45 or 7.5Hz) under different auditory conditions: no sound, /ba/, or /ki/ sounds. The frequency with which a shape was tagged was counterbalanced across participants. The sound was not frequency tagged, but presented at random (1sec) with 1-4sec intervals between sounds for the duration of visual stimulus presentation. Participants attended central fixation to detect a color change, or attended the shape to detect a border thickening. Signal-to-noise ratios for visual responses of the frequency tagged shapes were measured at the fundamental frequency at occipital (Oz, O1, O2),

frontal (AF3, F3, F4, AF4), and parietal (PO3, PO4) electrodes to quantify visual shape processing as a function of attention and audio-visual congruency. We found enhanced neural responses at occipital electrodes (1) for the attended shape when the sound was congruent vs incongruent - a crossmodal attention effect, (2) for the shape when it was attended versus unattended to attend fixation, despite the nature of the sound present (congruent vs incongruent) - a spatial attention effect. Overall, spatial attention effects were larger in the presence of congruent vs incongruent sounds. We did not find similar results at frontal or parietal electrodes. This work extends studies investigating mechanisms of attention by considering interactions between features across modalities for naturally occurring but abstract associations where it may not be necessary for features to be integrated into unitary objects.

Disclosures: E. Morina: None. **M. Beshara:** None. **V.M. Ciaramitaro:** None.

Poster

PSTR152. Cross-Modal Processing in Humans

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR152.26/DD3

Topic: D.08. Multisensory Integration

Support: UMass Boston undergraduate research funds

Title: Using frequency tagging to probe crossmodal correspondences: the influence of attention and audio-visual congruency on a neural signature of shape processing

Authors: *V. M. CIARAMITARO[1], E. MORINA[2], *V. M. CIARAMITARO[2];
[1]Psychology, Univ. of Massachusetts Boston - Boston, MA, Boston, MA; [2]Psychology, Univ. of Massachusetts, Boston, MA

Abstract: We used EEG frequency tagging with fast periodic visual and auditory stimulation to test (1) how attention influences shape processing, (2) how sounds influence shape processing, and (3) how visual & auditory stimuli interact. We studied crossmodal correspondences, specifically bouba-kiki associations, where abstract shapes, round vs spiky, are associated with nonsense sounds, /ba/ vs /ki/ respectively. This is an interesting model system since stimuli may be associated by naturally occurring statistical regularities or semantic associations, and may not be integrated the way we combine lip movements and the speech produced. Our paradigm involved presenting 3 stimuli simultaneously: 2 visual (round and/or spiky) and 1 auditory (/ba/, /ki/, or no sound) - each tagged at a unique frequency. Visual stimuli were contrast modulated at 5.45 or 7.5Hz, auditory stimuli were presented at 3Hz. Participants maintained central fixation and either attended fixation to detect color change, attended a shape to detect border thickening, or attended a sound to detect volume change. Our paradigm eliminated other ways in which stimuli could be associated: (1) temporal synchrony was absent - all stimuli changed at different frequencies and (2) the feature attended was irrelevant to the bouba-kiki association - participants attended shape thickness or sound loudness not shape spikiness or whether the sound

and shape matched. We hypothesized that frequency tagging would show (1) spatial attention effects: enhanced responses for an attended vs unattended shape and (2) crossmodal congruency effects: enhanced responses for a shape when audio-visual stimuli were congruent (eg, round shapes & /ba/ sounds) vs incongruent (eg, round shapes & /ki/ sounds). We also tested for crossmodal integration by measuring intermodulation frequencies. In a series of 3 experiments, a total of 97 adults, we computed signal-to-noise ratios for visual responses at the fundamental frequency of visual stimulus presentation at occipital (Oz, O1 & O2) electrodes to quantify shape processing. We found significant spatial attention effects: neural responses to the same shape were enhanced when attended vs ignored to attend fix or to attend another shape. We found no significant crossmodal congruency effect: neural responses to the same shape were not enhanced for congruent vs incongruent sounds (but see Morina et al., SFN 2023). We also found no evidence for audio-visual integration, no intermodulation frequency. This work extends studies investigating mechanisms of attention to features across modalities for abstract objects defined via associations.

Disclosures: V.M. Ciaramitaro: None. **E. Morina:** None. **V.M. Ciaramitaro:** None.

Poster

PSTR153. Cerebellum: Climbing Fibers

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR153.01/DD4

Topic: E.02. Cerebellum

Support: Wellcome Trust

Title: The genetic basis of functional subpopulations in the Inferior Olive

Authors: *E. GAGLIARDI, M. F. ZWART;
Univ. of St. Andrews, St. Andrews, United Kingdom

Abstract: Our ability to learn new movements and adapt them to an everchanging environment is an essential task for navigating the world. The inferior olivary nucleus (IO), which sits in our brainstem, is essential for this motor learning process. The IO is thought to detect mismatches between intended and actual movements; the IO then “teaches” the cerebellum by signalling the occurrence of movement errors to correct future movements. In spite of its importance, the exact function of the IO is still unclear. Due to its optical transparency, together with the ease of genetic and pharmacological manipulation, zebrafish represents a powerful model for in vivo imaging studies. Preliminary imaging evidence shows that IO cells have highly diverse neural activity profiles and encode selective representations of the sensory environment and motor output. The diversity in spatial location, dendritic morphology, axonal projection patterns and anatomically-defined classes of inferior olive neurons suggest a specific role of each cell subtype. How are these specified? We used scRNA-seq to disentangle the genetic complexity of the IO and delineate its single-cell expression profiles in 5 days old zebrafish, corroborated by

fluorescent in situ hybridisations to confirm the scRNA-seq data and reveal cluster marker genes. Candidate gene markers for each cluster were identified by MAST test for differential expression. We found a number of cell clusters in the Inferior Olive of 5 days old zebrafish and identified several differentially expressed genes. These newly acquired expression data will help to explore the neuronal diversity and functions of IO subtypes in more detail. Our study provides insight into the distinctive characteristics of the cells in the IO nucleus, allowing us to relate their genetic makeup to their activity patterns and motor learning to clarify their role in encoding sensory-motor errors.

Disclosures: **E. Gagliardi:** None. **M.F. Zwart:** None.

Poster

PSTR153. Cerebellum: Climbing Fibers

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR153.02/DD5

Topic: E.02. Cerebellum

Support: Wellcome Trust DBT India Alliance (500040/Z/09/Z and IA/S/17/2/503297)

Title: Developmental fine-tuning of cerebellar network and function in larval zebrafish

Authors: *S. VERMA, V. THIRUMALAI;
Neurobio., Natl. Ctr. for Biol. Sci., Bangalore, India

Abstract: The cerebellum, evolutionarily known for its role in motor control, undergoes massive ‘reorganization’ of neuronal connectivity during early postnatal development. Connections between cerebellar-Purkinje neurons (PNs) and Climbing fibers (CFs) are strongly implicated in sensory-motor feedback. CF-PN synapses are one of the strongest synapses in the brain and are known to undergo complex 'developmental pruning' to attain a one-to-one mapping (or mono-innervation). Previous studies in mammals have established a good understanding of the molecular and cellular pathways that orchestrate this process. In this study, we delve into the less understood physiological and behavioral relevance of cerebellar pruning using larval zebrafish. With the help of patch clamp electrophysiology, we first identify the timeline of mono-innervation of Purkinje neurons. We then couple electrophysiology with fictive opto-motor behavior assays to assess sensory-motor functions of CF-PN network. We do this by introducing feedback error in a simulated open loop landscape, thereby, perturbing larvae’s ergocentric spatial assessment. The salience of CF associated feedback error is enhanced as prevalence of mono-innervation increases over development. We also find that pruning of CF-PN synapses results in differently specialized populations of Purkinje neurons. These neurons can be sensory, motor or mixed depending on the specialization of the unique climber fiber associated with them. In summary, our study gives an insight into how a highly complex process of developmental

pruning specializes the cerebellar Purkinje neurons and enables them to compute and relay sensory-motor error signals with precision.

Disclosures: S. Verma: None. **V. Thirumalai:** None.

Poster

PSTR153. Cerebellum: Climbing Fibers

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR153.03/DD6

Topic: E.02. Cerebellum

Title: Zone-dependency of Purkinje cell dendritic Ca^{2+}dynamics originates from heterogeneous climbing fiber input

Authors: *S. KIM[1], S.-E. ROH[2], S. KIM[3], S. KIM[1];
[2]Dept. of Physiol., [1]Seoul Natl. Univ. Col. of Med., Seoul, Korea, Republic of; [3]Dept. of Physiol., Kyung Hee Univ. Col. of Oriental Med., Seoul, Korea, Republic of

Abstract: Climbing fiber (CF) input produces a complex spike in a Purkinje cell (PC), inducing strong depolarization with substantial Ca^{2+} influx in dendrites. Although the cerebellar circuitry was postulated to be uniform, it has recently been revealed that the cerebellar cortical circuit differs between compartmental zones separated by PC marker expressions such as Zebrin II. Hence, we set out to characterize the CF-evoked dendritic Ca^{2+} spike dynamics of PC between compartments in awake mice during resting and sensory processing. Also, we investigated the mechanism of differential PC responses between zones by directly observing CF Ca^{2+} activity. Surprisingly, GCaMP6f intensities appear to be greater in Zebrin II positive PC and this property enabled us to distinguish between different zones. CF-evoked PC Ca^{2+} spike frequency was lower and the amplitude/synchrony was higher in Zebrin II-positive (Z(+)) zones than in Zebrin II-negative (Z(-)) zones during the resting state. The sensory response of PC Ca^{2+} was distinct between zones: response probability and the amplitude/synchrony were prominent in Z(+) and slight in Z(-) zone. Interestingly, CFs projecting to Z(+) zones display enhanced amplitude/synchrony than CFs of Z(-) zones. Also, Z(+) CFs respond to sensory stimuli with high probability; Z(-) PC is almost not responding. The results suggest that presynaptic CF Ca^{2+} shapes zone-distinctive PC Ca^{2+} activity, sophisticating sensory processing.

Disclosures: S. Kim: None. **S. Roh:** None. **S. Kim:** None. **S. Kim:** None.

Poster

PSTR153. Cerebellum: Climbing Fibers

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR153.04/DD7

Topic: E.02. Cerebellum

Support: JSPS KAKENHI (JP19K07801, JP22K07324 to K.I., JP19K06883 to S.M., JP19H05208, JP19H05310, JP19K06882 to M.M., JP18H04012, JP20H05915, JP21H04785 to M.K., JP17H06313, JP22H05161, JP22H00460 to K.K.)
a Grant-in-Aid for Brain Mapping by the Integrated Neurotechnologies for Disease Studies (Brain/MINDS) (JP19dm0207079 to S.M., JP19dm0207080 to K.K.)
Fundacao para a Ciencia e a Tecnologia (PTDC/MED_NEU/32068/2017 to M.M.)
Takeda Science Foundation (to M.M. and K.K.)
the Uehara Memorial Foundation (to K.K.)

Title: Cerebellar climbing fibers jointly encode movement and reward signals during a voluntary forelimb movement task

Authors: *K. IKEZOE[1], N. HIDAKA[1], S. MANITA[1], M. MURAKAMI[1], S. TSUTSUMI[2], Y. ISOMURA[3], M. KANO[4], K. KITAMURA[1];
[1]Univ. of Yamanashi, Chuo, Yamanashi, Japan; [2]RIKEN Ctr. For Brain Sci., Wako, Saitama, Japan; [3]Tokyo Med. and Dent. Univ., Tokyo, Japan; [4]The Univ. of Tokyo, Tokyo, Japan

Abstract: Cerebellar climbing fibers (CFs) convey sensorimotor information and their error information, which are used for motor control and learning. Furthermore, they represent reward-related information. Despite such functional diversity of CF signals, it is still unclear whether the sensorimotor and reward-related information is conveyed by different CFs separately or by single CFs jointly. It is also unclear how the CFs encoding such different information are distributed over the cerebellar cortex. To address these issues, we performed two-photon calcium imaging from the dendrites of GCaMP6f expressing Purkinje cells (PCs) in the lobule V of the cerebellar vermis of adult mice (n = 8, all male), while the mice engaged in a voluntary forelimb lever-pull task. In this task, the mice were trained to pull a lever by their left forelimb and keep pulling it for 400 ms to obtain a water reward. We applied the spike deconvolution techniques to the dendritic fluorescence transients of PCs to estimate their complex spike timings. To characterize the responses of PCs during the task, we fitted the linear-nonlinear cascade encoding models to their deconvolved spike sequences with L2-regularization, which allowed us to avoid overfitting. The models contained five linear response kernels corresponding to five behavioral variables, namely lever position, speeds of lever pull and release, licking rate, and reward timing. We found that CF responses in 68% of 517 PCs could be explained significantly by the linear combination of the behavioral variables ($p < 0.05$, permutation test on estimated spike timings). The cluster analysis of the Gaussian mixture model clustering and the Bayesian information criterion showed that the PCs were classified into eight clusters according to the shapes of their response kernels. The responses of 81 (23%) PCs were modulated by both lever movement and reward delivery. Neighboring PCs exhibited similar CF response properties, forming functional clusters. The lateral width of the clusters was several hundred microns. Moreover, the functional clusters shared noise fluctuations of responses. Taken together, individual CFs convey multiple behavioral information in a multiplexed manner. The CFs are spatially organized into the

functional modules of the cerebellar cortex, which jointly encode sensorimotor and reward-related information. Preprint (bioRxiv) doi: https://doi.org/10.1101/2022.08.24.505210

Disclosures: **K. Ikezoe:** None. **N. Hidaka:** None. **S. Manita:** None. **M. Murakami:** None. **S. Tsutsumi:** None. **Y. Isomura:** None. **M. Kano:** None. **K. Kitamura:** None.

Poster

PSTR153. Cerebellum: Climbing Fibers

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR153.05/DD8

Topic: E.02. Cerebellum

Support: 18H05523
21H04810
JPMJSP2119

Title: Cerebellar Purkinje cell activity during temporal prediction of rhythmic visual stimulus

Authors: *L. LI, K.-I. OKADA, M. TANAKA;
Hokkaido Univ., Sapporo, Japan

Abstract: Accumulating evidence suggests that the cerebellum generates forward models in both motor and sensory processing. Previous studies in our laboratory have shown that neurons in the cerebellar dentate nucleus (DN) exhibit periodic activity when animals predict the timing of isochronously presented repetitive visual stimulus (Ohmae et al., 2013). Since pharmacological blockade of GABAergic input markedly reduces the periodic activity (Uematsu and Tanaka, 2022), activity in the DN may reflect the results of neural computation within the cerebellar cortex via inhibitory Purkinje cells (PCs). To understand the generation mechanism of temporal prediction signals, we examined the activity of PCs in behaving monkeys. Animals were trained to respond to the omission of periodically presented visual stimuli at an inter-stimulus interval (ISI) of 200, 400, or 600 ms. To detect stimulus omission, they needed to predict the timing of each repetitive stimulus. We have analyzed the activity of 51 well-isolated PCs recorded from the crus I and II lobules of three monkeys. Contrary to most DN neurons, more than half of PCs showed increased simple spike (SS) activity following each stimulus, consistent with their inhibitory nature. As with DN neurons, the modulation of SS activity after many repeats was proportional to the ISI and was greatly reduced when monkeys attempted to detect changes in color of the repetitive stimulus. Furthermore, the time course of periodic activity in individual DN neurons could be well explained by a weighted sum of signals from a subset of PCs, suggesting that the temporal prediction signals in the DN may originate primarily from the cerebellar cortex. The average firing rate of complex spikes (CSs) during each ISI remained unchanged, but CS tended to occur at specific times with stimulus repetition: in two-thirds of neurons, CS generation peaked 100-200 ms after the trough of SS activity. This appears to be consistent with the previous findings that CS causes long-term depression at parallel fiber

synapses that are active shortly (< 200 ms) before CS (Suvrathan et al., 2016). Indeed, after CS occurrence, the SS activity around the time of the next repetitive stimulus (± 50 ms) was reduced compared to that without a preceding CS (paired *t*-test, $p < 0.001$, n = 51), suggesting that the SS modulation might be shaped by CS through learning. Since cluster analysis of the time course of SS and CS activity can separate at least two groups of neurons, there may be multiple groups of PCs with different functions. Further investigation will reveal the generation mechanism of temporal signals within the local cerebellar circuitry.

Disclosures: L. Li: None. **K. Okada:** None. **M. Tanaka:** None.

Poster

PSTR153. Cerebellum: Climbing Fibers

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR153.06/DD9

Topic: E.02. Cerebellum

Support: 1R37NS128416

Title: Complex spike synchrony dissociates sensory from motor events

Authors: *J. PI, E. SEDAGHAT-NEJAD, M. FAKHARIAN, P. HAGE, R. SHADMEHR; Johns Hopkins Univ., Baltimore, MD

Abstract: Input from the inferior olive plays a major role in the function of the cerebellum, as suggested by near 100% chance of complex spike (CS) production in the post-synaptic Purkinje cell (P-cell). Theories of cerebellar function suggest that we learn how to move because the CS rates are modulated when movements are in error. However, CS rate changes are not specific to the erroneous movements. Rather, the CS rates change in response to a variety of sensory events and before a variety of movements. This diversity of events that modulate CS rates raises a problem for theories that view the cerebellum as a learning machine: how does the cerebellum differentiate sensory from motor events? We designed a 2x2 task in which a visual event was sometimes but not always followed by a movement, and a movement that was sometimes but not always preceded by a visual event. We then trained two marmoset monkeys (Callithrix jacchus) on a saccade task in which a primary visual target was jumped at saccade onset to a secondary location, inducing a motor error. We also had a task that had conditions in which visual events sometimes but not always instructed a movement, thereby trying to dissociate the encoding of sensory and motor events by complex spikes. During these tasks, we recorded, from oculomotor vermis lobules VI and VII, 389 individual P-cells and 289 simultaneously recorded pairs of P-cells. We found that each cell produced a CS at a preferred time following the visual input. When we sorted the cells based on the timing of their peak CS response rate we found that some P-cells produced their CS soon after the visual event, while others produced their CS immediately before the motor event. Following target onset, despite the sharp increase in the CS rates, there was little change in synchrony. In contrast, before saccade onset, there was both rise

in CS rates and synchrony. That is, the P-cells that produced their CSs following target onset were not synchronized with each other, but those that produced their CSs before saccade onset were. In summary, both visual and motor events modulated the CS rates across the population of P-cells. Thus, CS rates alone could not dissociate sensory from motor events. Some P-cells preferentially produced a CS following the visual event, while others produced a CS before the motor event. The CSs following the visual event were not temporally synchronized. In contrast, the CSs before the motor event were synchronized. As a result, each P-cell received an olivary input that specialized in conveying either sensory or motor events. The olivary cells that conveyed motor events did so synchronously. As a result, CS synchrony, and not CS rates, dissociated between sensory and motor events.

Disclosures: J. Pi: None. **E. Sedaghat-Nejad:** None. **M. Fakharian:** None. **P. Hage:** None. **R. Shadmehr:** None.

Poster

PSTR153. Cerebellum: Climbing Fibers

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR153.07/DD10

Topic: E.02. Cerebellum

Title: Error Detection by the Olivocerebellar System and its Role in Motor Adaptation

Authors: *H. SHAWEIS, M. ZWART, P. N. MULLEN;
Sch. of Psychology and Neurosci., Univ. of St. Andrews, St Andrews, United Kingdom

Abstract: The ability to learn and refine movements is crucial for adapting to an ever-changing environment. The inferior olive is believed to play a key role in this learning process by detecting mismatches between predicted and actual sensory feedback during movements. However, the encoding of sensory information in the inferior olive remains unclear. To investigate this, we conducted two-photon calcium imaging of inferior olive neurons expressing GCaMP6f in 5-7dpf zebrafish larvae. We presented different sensory modalities, including visual, flow, and motion stimuli, and generated distinct neuronal maps for each stimulus by regressing neuronal responses with sensory regressors. We quantified directional tuning by computing the vector sum of neuronal responses to eight different stimulus directions. Neurons were ranked based on t-scores and slope from regression analyses, with the 95th percentile considered responsive to a specific sensory modality. Strong responses were observed for flow and motion stimuli along the head-tail (anterior-posterior) axis. Interestingly, all visual stimulus directions elicited robust responses, revealing a highly organized spatial representation and selective tuning of inferior olive neurons to visual input. Like flow and motion modalities, the largest visual responses were observed in the head-tail direction. There was a modest convergence between sensory modalities (8-12%). Simultaneous electrophysiological recordings of motor activity from the tail showed increased fictive swim bouts in response to forward moving visual stimuli. Neurons correlated with this motor onset also responded to forward

moving visual stimuli in the absence of behavior. These findings suggest that sensory information is highly organized in the inferior olive and that neurons have a bias towards those stimuli that reflect the zebrafish's position along the anterior-posterior axis in space.

Disclosures: H. Shaweis: None. **M. Zwart:** None. **P.N. Mullen:** None.

Poster

PSTR153. Cerebellum: Climbing Fibers

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR153.08/DD11

Topic: E.02. Cerebellum

Title: Climbing fiber input continuously maintains, rather than permanently shapes, the simple spike output of Purkinje cells

Authors: *S. Z. MULLER[1], R. SHADMEHR[2], L. ABBOTT[1], N. SAWTELL[1];
[1]Columbia Univ., New York, NY; [2]Dept Biomed. Eng, Johns Hopkins Univ. Dept. of Biomed. Engin., Baltimore, MD

Abstract: A dominant view in the field of cerebellar studies is that climbing fiber input to Purkinje cells serves as an error signal conveying information about deviations from a desired state. Similar to supervised learning in artificial neural networks, learning in the cerebellum is assumed to proceed until error is eliminated and then stop. Although this view has been contested on several grounds, a large body of evidence from studies of various forms of eye movements and classical conditioning is widely held to support it. Here we argue based on careful review of eye movement and classical conditioning studies that the elimination of error is not sufficient to maintain learned changes in the simple spike firing of Purkinje cells. In fact, in all cases, complex spike firing occurs prior to eye movements even when no error occurs. Furthermore, stability analysis of climbing fiber driven plasticity at parallel fiber synapses shows that some form of decay of synaptic weights to baseline values is needed to achieve equilibrium. Thus, we conclude that maintenance of a behavior that depends on climbing fiber input requires continuous modulation of complex spike firing to shape the desired simple spike output.

Disclosures: S.Z. Muller: None. **R. Shadmehr:** None. **L. Abbott:** None. **N. Sawtell:** None.

Poster

PSTR153. Cerebellum: Climbing Fibers

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR153.09/DD12

Topic: E.02. Cerebellum

Support: NIH grant R34 NS118445

Title: The Role of Neocortico-Olivo-Cerebellar Pathway in Cerebellar Prediction of Neocortical Signals

Authors: K. OHMAE, Z. S. HASSAN, J. F. MEDINA, ***S. OHMAE**;
Baylor Col. of Med., Houston, TX

Abstract: The synergistic collaboration between the neocortex and cerebellum facilitates sophisticated information and language processing. The cerebellum is hypothesized to predict neocortical outputs to promote neocortical processing and enhance processing speed and accuracy. A growing body of evidence supports this hypothesis (Ito, 2008; Li & Mrsic-Flogel, 2020; Wagner & Luo, 2020). However, the mechanisms underlying the cerebellum's acquisition of this prediction are yet to be elucidated. Based on the anatomical understanding that the inferior olive, the origin of prediction error signals crucial for cerebellar prediction function, receives substantial disynaptic projections from the neocortex, it is hypothesized that the cerebellum learns to predict neocortical signals through the neocortico-olivo-cerebellar pathway (Ito, 2008; Inoue et al., 2016; Sokolov et al., 2017). This study aims to validate this hypothesis by investigating whether Purkinje cells in the cerebellum can receive neocortical signals through the neocortico-olivo-cerebellar pathway and learn to make predictions. We conducted experiments utilizing electrical stimulation to elicit signals in the periocular sensory areas of the mouse sensory neocortex after a visual cue. Our objectives were twofold: (1) to determine if Purkinje cells in the eyeblink control region of the cerebellum receive the neocortical signal transmitted via the inferior olive, and (2) to ascertain if these same cells learn to generate a preceding signal to predict the neocortical signal. We found that Purkinje cells receive the neocortical signal through the inferior olive in less than 15 milliseconds. Furthermore, these cells learned to produce predictive outputs associated with eyeblink movements to protect the eye. Notably, when the cerebellum successfully predicted the neocortical signal, the neocortico-olivo-cerebellar signal was suppressed (i.e., prediction error of neocortical signal = neocortical signal - cerebellar prediction). These results provide compelling evidence that the cerebellum can learn to predict the neocortical signals and receives the errors of this prediction via the inferior olive.

Disclosures: K. Ohmae: None. **Z.S. Hassan:** None. **J.F. Medina:** None. **S. Ohmae:** None.

Poster

PSTR153. Cerebellum: Climbing Fibers

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR153.10/DD13

Topic: E.02. Cerebellum

Support: NIH Grant NS124217

Title: The climbing fiber provides an instructive signal for plasticity outside the cerebellum

Authors: A. M. SILBAUGH[1], *C. HANSEL[2];
[1]Univ. of Chicago, [2]Neurobio., Univ. of Chicago, Chicago, IL

Abstract: The classic example of supervised learning occurs at the parallel fiber (PF) to Purkinje cell synapse in the cerebellum, where plasticity depends on co-activity of the climbing fiber (CF) input, which - according to the Marr-Albus-Ito theory - provides an instructive signal. This well-studied CF function within the cerebellar system is in stark contrast to what is known about the potential relevance of CF activity outside of the cerebellum. A plausible anatomical pathway for interactions with neocortical areas has been described, which includes activation of Purkinje cells, and the subsequent signal transfer via the cerebellar nuclei and relay nuclei in the thalamus, in particular the ventral lateral (VL) and the posteriormedial (Pom) nuclei. Here, we asked whether CF co-activity can provide an instructive signal that affects receptive field (RF) plasticity in the primary somatosensory (S1; barrel) cortex of mice. Using intrinsic optical imaging, we show that repetitive activation of individual whiskers (via attached glass capillaries; 5min @ 10Hz) enhances whisker representation in the barrel cortex (N=4 mice). In control experiments (no whisker 'tetanization'), the intrinsic signal remains stable (N=4 mice). Optogenetic co-activation of channelrhodopsin 2 (ChR2)-expressing CFs in the cerebellum suppresses this form of cortical RF plasticity (N=4 mice). We used two-photon imaging from awake, adult mice to obtain access to specific neuron populations. In particular, we focused on GCaMP6f-based calcium measurements from L2/3 pyramidal cells and PV+ interneurons (the latter are preferentially activated by single whisker stimuli co-applied with optogenetic CF activation). PV+ interneurons are identified by expression of tdTomato. Pyramidal neurons are identified by morphological shape. To obtain tdTomato labeling of PV+ interneurons, all experiments are performed using PV-Cre mice with a Cre-dependent tdTomato AAV (AAV9-FLEX-tdTomato). For imaging, we use the GCaMP vector AAV9.Syn.GCaMP6f.WRPE.SV40, which labels all neurons. As in the intrinsic imaging experiments, whisker 'tetanization' (5min @ 10Hz) caused RF plasticity, now observed as an increase in response amplitude in a subgroup of L2/3 pyramidal cells (n=125 neurons; N=4 mice; no effect on PV+ cells). CF co-activation suppressed RF plasticity in L2/3 pyramidal cells (n=497 neurons; N=5 mice). These data show that CF activity in the cerebellum may provide an instructive signal to regulate cortical RF plasticity, possibly by recruiting activity in PV+ interneurons.

Disclosures: A.M. Silbaugh: None. **C. Hansel:** None.

Poster

PSTR154. Basal Ganglia: Physiology and Function

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR154.01/DD14

Topic: E.03. Basal Ganglia

Support: R01 NS091144/NS/NINDS NIH HHS/United States

Title: Motor learning induces dynamic reorganization of the activity and structure of primary motor cortical axonal boutons in the striatum

Authors: *M. SHENG[1], D. LU[2], J. DING[2];
[1]Neurosurg., Stanford Univ., Stanford, CA; [2]Neurosurg., Stanford University, Sch. of Medicine, Wu Tsai Neurosci. Inst., Stanford, CA

Abstract: The primary motor cortex (M1) and dorsal lateral striatum (DLS) are required for the learning and execution of motor skills. Synaptic plasticity of corticostratial synapses is pivotal for acquisition of motor skills, but mechanisms remain unclear. Here, we repeatedly imaged the activity and structural dynamics of the same M1 axons in the DLS while the mice were trained to learn a cued-lever pushing motor task using *in vivo* 2-photon microscopy. We find that the activities of M1 axons become stereotyped after motor learning, and the pairwise trial-to-trial activity patterns show an increase in cross-correlation when performing a similar lever pushing motion at the late phase of learning. Interestingly, we find that the activities of M1 boutons exhibit selectivity for rewarded movements (RM) and un-rewarded movement (UM), and motor learning significantly increased the fraction of RM boutons and decreased that of UM boutons. To investigate what caused such a shift, we repeated imaged the same M1 axonal segment in the DLS and quantified the formation and elimination of axonal boutons. We find that motor learning induced an increase in formation of new axonal boutons followed by an increase in pruning of the boutons in DLS. Furthermore, the newly formed axonal boutons become more stabilized and are clustered along axonal segments. Together, these results demonstrate that learning of a novel motor skill involves both structural and functional reorganization of M1 axonal boutons, leading to the formation and stabilization of the ensemble of M1 axonal outputs to DLS, which may be critical for the learning and execution of learned movements.

Disclosures: **M. Sheng:** None. **D. Lu:** None. **J. Ding:** None.

Poster

PSTR154. Basal Ganglia: Physiology and Function

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR154.02/DD15

Topic: E.03. Basal Ganglia

Support: McKnight Foundation

Title: Neuron-dependent Auditory Feedback in the Zebra Finch

Authors: *J. SCHERRER[1], M. S. FEE[2];
[1]MIT, Cambridge, MA; [2]Brain & Cog Sci. / McGovern Inst., Massachusetts Inst. Tech., Cambridge, MA

Abstract: The songbird Zebra Finch (Taeniopygia castanotis) learns a complex motor sequence through a trial-and-error process suggestive of reinforcement learning. This learning process

requires a basal ganglia-thalamocortical loop called the anterior forebrain pathway (AFP). Existing evidence suggests that the AFP learns a time-dependent bias signal that steers the motor pathway to avoid vocal errors, and this bias signal is known to be dependent on the cortical output of the AFP known as LMAN (lateral magnocellular nucleus of the anterior nidopallium). However, little is known about the neural code in LMAN that underlies this bias signal, or how this neural code is learned and generated. We seek to address these questions through a variant of the conditional auditory feedback (CAF) paradigm in which noise bursts are played to the bird during song to disrupt its performance evaluation conditional on parameters of the song. In our experiments, these noise bursts are instead played contingent on the calcium activity of individual neurons in LMAN recorded in real time using an extremely lightweight head-mounted microscope (neuron-contingent CAF or nCAF). We present preliminary data on the effects of nCAF on neural activity patterns in LMAN and their implications for understanding the function of the AFP.

Disclosures: **J. Scherrer:** F. Consulting Fees (e.g., advisory boards); Open Ephys. **M.S. Fee:** None.

Poster

PSTR154. Basal Ganglia: Physiology and Function

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR154.03/DD16

Topic: E.03. Basal Ganglia

Support: KAKENHI (JP22H05157 and JP23H02781 to K.I, JP19H05467 to M.T)
AMED (JP20dm0307021 to K.I., and JP21dm0207077 to M.T)

Title: Parvalbumin-positive neuron-selective gene transduction into the monkey striatum

Authors: *A. ZHENG[1], K. KIMURA[1], Y. NAGAI[2], M. FUJIWARA[1], M. NAKANO[1], K. NAGAYASU[2], K.-I. INOUE[1], M. TAKADA[1];
[1]Ctr. for the Evolutionary Origins of Human Behavior, Kyoto Univ., Inuyama / Aichi Prefecture, Japan; [2]Dept. of Mol. Pharmacology, Grad. Sch. of Pharmaceut. Sci., Kyoto Univ., Kyoto, Japan

Abstract: Nonhuman primates have widely been utilized as invaluable models for studying the neural mechanisms underlying a variety of higher brain functions by virtue of their significant relevance to humans. To investigate the causal relationship between a specific neural circuit and its corresponding behavioral property, it is essential to elucidate the functional roles of distinct types of neurons within the circuit, which would be effectively and efficiently achieved by neuron type-selective gene manipulation. However, only a limited number of reports have been available on selective targeting of neuron types in nonhuman primates, because experimental techniques have poorly been developed for delivering functional molecules, such as opsins and designed receptors, into a genetically-identified neuronal population. Therefore, improvement in novel tools that allow neuron type-selective gene transduction would provide us with a

breakthrough of research approaches to understanding of higher brain functions in nonhuman primates. Parvalbumin-positive (PV+) neurons in the striatum, known as fast-spiking interneurons, have been implicated in motor control, reinforcement learning, and decision-making, despite that they constitute only less than 1% of the total striatal neurons, including both projection neurons and interneurons. In nonhuman primates, however, the exact functional role of these PV+ neurons remains unclear due to difficulty in targeting such a sparsely-distributed neuronal population. In the present study, we isolated several PV promoter candidates of different lengths from the conserved PV promoter region in the macaque genome. Through initial screening using lentiviral vectors in mice, we identified two promising candidates, PV0.8 and PV1.8, which showed superior neuron type-specificity via gene transduction. Subsequently, we injected adeno-associated virus (AAV) vector carrying PV0.8 or PV1.8 into the striatum of macaque monkeys, and assessed the neuron type-selectivity of transgene expression using double immunostaining for PV and the reporter gene GFP. Remarkably, both candidates exhibited a high selectivity as evidenced by the fact that over 80% of GFP-expressing neurons were co-localized with PV. This result indicates that our innovative PV-specific AAV vectors with the macaque promoter may offer an excellent methodology to deliver functional molecules selectively into PV+ neurons in the monkey striatum for exploration of their contributions to striatum-related neural circuit functions.

Disclosures: **A. Zheng:** None. **K. Kimura:** None. **Y. Nagai:** None. **M. Fujiwara:** None. **M. Nakano:** None. **K. Nagayasu:** None. **K. Inoue:** None. **M. Takada:** None.

Poster

PSTR154. Basal Ganglia: Physiology and Function

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR154.04/DD17

Topic: E.03. Basal Ganglia

Support: NIH Grant NS094184
NIH Grant NS113922
NSF Neuronex 2014862

Title: Synaptic integration of somatosensory and motor cortical inputs onto medium spiny neurons of the caudate putamen.

Authors: *V. SAMPATHKUMAR[1,2], K. KOSTER[1], B. J. CARROLL[1], S. SHERMAN[1], N. B. KASTHURI[1,2];
[1]Neurobio., Univ. of Chicago, Chicago, IL; [2]Biosci. Div., Argonne Natl. Lab., Lemont, IL

Abstract: The basal ganglia plays a pivotal role in various brain functions, including motor control and cognitive functioning. We focus here on the basal ganglia region innervated by layer 5 (L5) of cortex, namely the caudoputamen (CP). The most abundant CP cell type is the GABAergic medium spiny projection neuron (MSN), whose spines are the primary targets of the

cortical input. It is known that projections from different cortical regions overlap in the CP, but it remains unknown whether inputs from different cortical areas converge at the level of individual MSNs. To address this, we employed genetic labeling to distinguish L5 neurons from somatosensory (S1) and motor (M1) cortices in electron microscopic (EM) data sets and combined it with large volume serial EM. We find convergence of L5 S1 and M1 cortical inputs onto reconstructed MSNs. These inputs had terminal diameters ranging from 0.4-1.4 μm, often contained mitochondria, and were predominantly located on proximal dendrites. We observed more synapses from M1 than from S1. The cortical inputs primarily targeted spines of MSNs with <5% onto dendritic shafts of aspiny inhibitory interneurons. Of the targeted spines, most had a spine apparatus, an ultrastructural indication of strong synapses. To gain further insight into how cortical outputs influence circuit dynamics, we used fluorescence microscopy to identify regions of CP where L5 S1 and M1 terminals overlapped the most. Then, using optogenetics in an acute slice preparation, we demonstrated a depressing paired-pulse synaptic response resulting from activation of either S1 or M1 inputs onto physiologically identified MSNs, consistent with both inputs being drivers. These findings suggest that individual MSNs serve as substrates of sensorimotor integration, which is passed on as an inhibitory input to the output nucleus of the basal ganglia, the internal segment of the globus pallidus, to provide a strong inhibitory and thus presumably gating input to motor thalamus. Thus, these layer 5 inputs from cortex may integrate to control the gating properties exhibited by the basal ganglia onto motor thalamus.

Disclosures: V. Sampathkumar: None. **K. Koster:** None. **B.J. Carroll:** None. **S. Sherman:** None. **N.B. Kasthuri:** None.

Poster

PSTR154. Basal Ganglia: Physiology and Function

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR154.05/DD18

Topic: E.03. Basal Ganglia

Support: the Israel Science Foundation
German Collaborative Research Center TRR295 (Returning dynamic motor network disorders using neuromodulation)
Israel-china bi-national science foundation

Title: Neural synchronization: a shared mechanism in dissociative and hypnotic sedation

Authors: *J. GUANG[1], H. BAKER[2], O. BEN-YISHAY NIZRI[2], S. FIRMAN[3], U. WERNER-REISS[5], V. KAPULLER[4], Z. ISRAEL[6], H. BERGMAN[2];
[1]Hebrew Univ. of Jerusalem, Jerusalem, Israel; [2]Hebrew Univ., [3]Dept. of Anesthesiol., [4]Dept. of Pediatric Surgery, Hadassah Med. Ctr., Hebrew Univ., Jerusalem, Israel; [5]The Hebrew Univ. of Jerusalem, The Hebrew Univ. - Hadassah Med. Sch., Jerusalem, Israel; [6]Hadassah Hosp., Hadassah Hosp., Jerusalem, Israel

Abstract: Accurate placement of microelectrode is the crucial part of deep brain stimulation (DBS) treatment for Parkinson's disease (PD). It heavily depends on the activity of landmark brain regions, such as subthalamic nucleus (STN) and globus pallidus internal segment (GPi). Patients are often required to keep awake during this procedure since the effects of anesthesia drugs on the physiology of those landmark brain regions are not well-studied. To address this question, we recorded EEG and local filed potential (LFP) from the external globus pallidus (GPe), the central nucleus of the basal ganglia, which is connected with both STN and GPi, of two female African green monkeys before, during, and after a moderate sedation with five commonly used drugs, ketamine, propofol, remifentanil, dexmedetomidine, and scopolamine. The dose of each drug was determined by titration via evaluations of eye-close, vital signs, electro-oculogram (EOG) and electromyography (EMG). Classical spectral analysis, Fitting Oscillation and One Over F (FOOOF) analysis and pairwise correlations were used to study the effects of sedation drugs on brain areas' activities. We found that ketamine increases power in high beta/gamma band and decrease in total power, and for other sedation drugs we saw increase in delta/theta power, decrease in low beta power and an increase in the total power. Moreover, all the sedation drugs had pairs that show synchronization increase. The dynamics of changes in spectrums were faster than the dynamics of changes in correlations. And the results from four different analysis methods, classical and FOOOF spectrum, auto and pairwise correlation had the same trend. Our findings shed light on the neural mechanism of sedation and might serve as a reference for sedation drug selection on microelectrode placement surgery for PD patients.

Disclosures: J. Guang: None. **H. Baker:** None. **O. Ben-Yishay Nizri:** None. **S. Firman:** None. **U. Werner-Reiss:** None. **V. Kapuller:** None. **Z. Israel:** None. **H. Bergman:** None.

Poster

PSTR154. Basal Ganglia: Physiology and Function

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR154.06/DD19

Topic: E.03. Basal Ganglia
NIH DA R01 027222

Title: Acute and chronic treatment of methylphenidate induced sensitization and cross-sensitization in a female animal model of ADHD

Authors: E. J. LAM[1], A. C. SWANN[2], *P. DASH[2], N. DAFNY[2];
[1]Univ. of Texas Med. Sch. at Houston, Houston, Texas, Houston, TX; [2]Univ. of Texas Med. Sch. at Houston, Houston, TX

Abstract: Acute and chronic treatment of methylphenidate induced sensitization and cross-sensitization in a female animal model of ADHD
E. J. Lam, A. C. Swann, P. Dash* and N. Dafny
Department of Neurobiology and Anatomy, The University of Texas McGovern Medical School at Houston, 6431 Fannin Street, Houston, TX 77030, USA

Psychostimulants, such as methylphenidate(MPD) and amphetamine (AMPH), are commonly prescribed medications for individuals afflicted with attention deficit hyperactivity disorder (ADHD). Yet, the full effects of these medications on individuals with ADHD are not clearly understood and need further elucidation. The objectives of the study were (1) to examine whether acute and chronic treatment of MPD induced sensitization and (2) to ascertain whether prior exposure to MPD could cause cross-sensitization to AMPH in female Spontaneously Hypertensive/Hyperactive Rat (SHR) strain, an animal model of ADHD, compared to normal Wistar-Kyoto (WKY) and Sprague-Dawley (SD) strains. Different regimens of MPD and AMPH (i.p) involving 24 groups of female WKY, SHR, and SD rats (each, n = 8) were used. Locomotor behavior activity was recorded for 2-hour before and post-injection of saline, MPD, and AMPH during their adolescence and adulthood. MPD (0.6, 2.5 &10.0mg/kg) and AMPH (0.6mg/kg) elicit increase in TD traveling, Chronic MPD and AMPH elicit further significant ($p<0.05$) increases in TD traveling as compared to their initial effects, i.e., chronic MPD or AMPH elicit behavioral sensitization. MPD or AMPH treatment to these behavioral sensitized groups result in further significant ($p<0.05$) increase in TD activity, i.e., cross sensitization were observed. Sensitization and cross sensitization are an experimental marker indicating that the drugs are causing dependent and are addicted, suggesting that both drugs are causing dependent and addiction. Support in part by NIH DA R01 027222.

Disclosures: E.J. Lam: None. **A.C. Swann:** None. **P. Dash:** None. **N. Dafny:** None.

Poster

PSTR154. Basal Ganglia: Physiology and Function

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR154.07/DD20

Topic: E.03. Basal Ganglia

Support: BRAIN Initiative NINDS R00 R00NS112417
American Parkinson's Disease Association (APDA)
2021APDA00RG00000209666

Title: Functional effects of exercise and cholinergic neuron manipulation in the pedunculopontine nucleus

Authors: *C. B. SCOTT[1], Z. COLON[2], M. R. CROOM[2], R. C. EVANS[3];
[1]Georgetown Univ. Med. Ctr., [2]Georgetown Univ. Med. Ctr., Washington, DC; [3]Georgetown Univ. Med. Ctr. Interdisciplinary Program In Neurosci., Washington, DC

Abstract: Exercise is neuroprotective in various aging-related diseases such as Parkinson's Disease (PD). In PD, motor learning is significantly impacted (Rinne et al., 2008). Importantly, exercise improves motor learning in both rodents and humans and decreases motor impairment severity once the disease has already progressed (Li & Spitzer, 2020; Wanner et al., 2021). To explore exercise as a protective measure, it is important to understand how neural circuits are

affected. A key brain structure modified by exercise is the pedunculopontine nucleus (PPN). Cholinergic neurons of the PPN have been implicated in motor skill acquisition and the loss of these neurons impairs motor learning (MacLaren et al., 2014). To isolate the neural circuitry that connects exercise and motor learning, we gave mice free access to a running wheel in their home cages. After a week of exercise, we assessed motor skill acquisition and retention via an accelerating rotarod assay and balance beam training. Using both sexes of wild-type C57BL/6J mice, we found those with running wheel access performed significantly better than control animals. Since the cholinergic PPN neurons are implicated in enhanced motor learning we evaluated their characteristics through the following approaches: ex vivo whole cell patch clamp electrophysiology, optogenetics, and acute versus chronic chemogenetics. We recorded the electrical activity of PPN neurons in ChAT-cre/tdTomato mice and found the amplitude of spontaneous excitatory postsynaptic currents (sEPSC) were significantly increased post-exercise. Since exercise enhances learning and increases the excitatory synaptic input to the cholinergic PPN, we tested whether direct excitation of these neurons would enhance motor learning without exercise. To directly excite the cholinergic PPN neurons, we used optogenetics or designer receptors exclusively activated by designer drugs (DREADDs) in ChAT-cre mice. We found that acute excitation of the cholinergic neurons did not improve motor skill acquisition. However, inhibition of the cholinergic neurons impaired early motor learning. To follow up on these results, we propose chronic administration of a DREADD agonist to supplement the acute results. We hypothesize that chronic excitation of the cholinergic PPN will enhance motor learning while chronic inhibition of these neurons will impair motor learning. Understanding how cholinergic PPN neurons contribute to the early stage of motor learning and are altered by exercise will yield important insight into how exercise alters brain circuitry to improve learning.

Disclosures: C.B. Scott: None. **Z. Colon:** None. **M.R. Croom:** None. **R.C. Evans:** None.

Poster

PSTR154. Basal Ganglia: Physiology and Function

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR154.08/DD21

Topic: E.03. Basal Ganglia

Support: ANR-18-CE37-0018
AMX-19-IET-004
ANR-17- EURE-0029

Title: Sub-cortical activity patterns in reward-associated modulation of motor adaptation

Authors: *K. PUNJABI[1,2], P. TILSLEY[3], B. NAZARIN[1], J. SEIN[1], M. WEYGANT[4,5], F. SARLEGNA[6], N. MALFAIT[1], J.-P. RANJEVA[3], J.-P. STELLMANN[3,7];
[1]INT UMR 7289, [2]Crmbm umr 7339, [3]CRMBM UMR 7339, Aix-Marseille Univ., MARSEILLE, France; [4]NeuroCure Clin. Res. Ctr., Charité – Universitätsmedizin Berlin, Berlin, Germany; [5]Exptl. and Clin. Res. Ctr., Max Delbrueck Ctr. for Mol. Med. and Charité –

Universitätsmedizin, Berlin, Germany; [6]ISM UMR 7287, CNRS and Aix-Marseille Univ., MARSEILLE, France; [7]Dept. of Neuroradiologie, APHM, Timone Univ. Hosp., MARSEILLE, France

Abstract: Adapting to delays in visual feedback of movements is increasingly common with the use of digital devices and is impacted by reward at the end of movement. Classical motor control theories assign reward-based learning and timing motor commands to the basal ganglia (BG), and sensory prediction error (SPE)-based learning to the cerebellum (Cb), and their feedbacks' interacting at the cortex. Recent evidence supports Cb's involvement in reward processing and direct subcortical connections to BG. Human motor adaptation studies examine behavior and electrophysiology, missing subcortical interactions which can be studied using functional Magnetic Resonance Imaging (fMRI). While 3T is the standard, 7T fMRI offers higher statistical power in detecting small effects. We investigate BG-Cb activity patterns associated with prediction error (RPE) and SPE in 3T and 7T using a block-event mixed paradigm.
Participants intercepted a target moving on a screen using ballistic movements with a joystick. RPE was studied through manipulated outcome feedback, rewarding 50% successful trials with audio-visual explosion. SPE was studied by introducing a lag between joystick position and its visual feedback. Three trial types (R+, R-, M) and two block types (Baseline, Delay) were defined. The task consisted of two rounds of consecutive baseline and delay blocks. Five adults (2 F; mean age 26.4 ± 1.9 yrs) practiced the task and underwent fMRI at 3T and 7T on days 1, 3 and 5, with counterbalanced scan order. Anatomical and functional images were preprocessed with fMRIprep. A 1st level General Linear Model (GLM) design included onsets of trials and blocks, and nuisance regressors. Contrasts were computed for each participant: R- vs M (success vs failure), R+ vs R- (reward perception), and delay vs baseline (sensorimotor adaptation). One-sample t-tests were conducted in SPM12 for group-level analysis on normalized contrasts. Using the subcortical nuclei atlas (2018) and Diedrichson cerebellar atlas (2009), activation clusters in BG and Cb ROIs were inspected for cluster peak location differences between 3T and 7T.
Results revealed distinct peak activations in BG and Cb for all three contrasts at both 3T and 7T at similar thresholds. Lateralization changes in putamen and Cb clusters were observed between the two fields for the success vs failure contrast.
Using kinematic behaviour-informed analysis, we are exploring if these results imply that the difference between the fields extends beyond signal power, and if 7T fMRI is a better tool to precisely resolve individualised brain networks resulting from inter-personal differences in choice of adaptation strategy.

Disclosures: **K. Punjabi:** None. **P. Tilsley:** None. **B. Nazarin:** None. **J. Sein:** None. **M. Weygant:** None. **F. Sarlegna:** None. **N. Malfait:** None. **J. Ranjeva:** None. **J. Stellmann:** None.

Poster

PSTR154. Basal Ganglia: Physiology and Function

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR154.09/DD22

Topic: E.03. Basal Ganglia

Support: Maharjan Thesis Grant
NIH Grant 1U19N5123716
NIH Grant RF1MH126883

Title: Roles of direct and indirect pathway neurons during auditory decision-making

Authors: *D. M. MAHARJAN[1], A. ZADOR[2];
[1]Cold Spring Harbor Lab., Huntington, NY; [2]Cold Spring Harbor Lab., Cold Spring Harbor, NY

Abstract: Understanding how the brain processes sensory information and converts it into motor responses remains a crucial question in neuroscience. To study the formation and exploitation of such sensory-motor associations in the brain, our laboratory has developed an auditory two-alternative forced choice task called the 'cloud of tones' task. In this task, rodents are trained to associate making left or right movements with the presentation of auditory stimuli containing either a high frequency cloud of tones or a low frequency cloud of tones. Selective activation of corticostriatal neurons - those projecting from auditory cortex to auditory striatum - shifts the perception of stimulus content in rodents, biasing choices in a manner predicted by the tonotopy of the corticostriatal projections (Znamenskiy, Nature, 2013). Furthermore, the formation of these learned associations results in synaptic strengthening of these corticostriatal neurons specifically onto auditory striatum (Xiong, Nature, 2015). Neurons in the auditory striatum primarily consist of direct pathway (D1) and indirect pathway (D2) medium spiny neurons, two cell-types postulated to have opposing effects on motor behavior. Here, we investigate the involvement of D1 and D2 neurons within the auditory striatum in regulating auditory decision-making. Using an optogenetic approach to artificially manipulate the activity of D1 and D2 neurons, we provide evidence that these two cell-types in the auditory striatum circuitry play a crucial role in modulating this behavior. We unilaterally activated channelrhodopsin-2 expressing D1 or D2 neurons in 10% of the trials by illuminating blue light through implanted fibers. Activation of D1 neurons led to a significant behavioral bias towards the contralateral direction. Conversely, activation of D2 neurons resulted in a bias towards the ipsilateral direction. We then conducted fiber photometry recordings of GCaMP8f expressing D1 or D2 neurons while animals actively performed the cloud of tones task. Interestingly, our observations indicate that D1 neurons are specifically active during task performance, whereas D2 neurons were significantly less active. We find that the responsivity of D1 activity is influenced by the acoustic characteristics of the auditory stimuli. Our results suggest that D1 activity, not D2 activity, regulates auditory decision-making by aiding in the discrimination of auditory stimulus content.

Disclosures: D.M. Maharjan: None. **A. Zador:** None.

Poster

PSTR154. Basal Ganglia: Physiology and Function

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR154.10/DD23

Topic: E.03. Basal Ganglia

Support: NIH NIDA Grant K00DA058542
NIH Grant MH124004
NIH Shared Instrumentation Grant S10OD020039

Title: Striatal Association Megaclusters

Authors: *H. L. KOSAKOWSKI, N. SAADON-GROSMAN, R. BUCKNER;
Harvard Univ., Cambridge, MA

Abstract: The striatum receives projections from multiple regions of cerebral cortex consistent with its role in diverse sensorimotor, affective, and cognitive functions. Monkey anatomical connectivity shows that the dorsomedial striatum (i.e., caudate) receives input from frontal and parietal regions of association cortex (Selemon & Goldman-Rakic 1985; Haber et al. 2006). An open question is how inputs to the caudate from higher-order cortical zones are organized. Recent advances in precision neuroimaging indicate that at least five interdigitated networks populate human association cortex (abbreviated DN-A, DN-B, LANG, FPN-A, and FPN-B). Within the cerebral cortex, these five networks are organized with repeating adjacencies forming clusters across multiple association zones. We refer to these as Supra-Areal Association Megaclusters (SAAMs). Here, we used precision neuroimaging techniques to map the complex organization of the striatum open to the possibility that the caudate may recapitulate the organization of association networks found in the cerebral cortex. First, we adopted a winner-takes-all parcellation strategy using functional connectivity on resting-state fMRI data (adapted from Choi et al. 2011). The approach was developed on two intensely studied pilot participants (Xue et al. 2021) and then prospectively applied to 15 new participants. In most individuals, we observed adjacent regions of caudate that were correlated with five separate cerebral networks – DN-A, DN-B, LANG, FPN-A, and FPN-B – recapitulating the SAAMs. To further interrogate this organization, we placed seed regions in the separate striatal regions within each individual and observed correlations in cerebral cortex that adhered to network boundaries. Taken together, these results indicate that the caudate possesses regions preferentially associated with multiple distinct association networks, extending the general notion of parallel specialized basal ganglia circuits (Alexander, Delong, & Strick 1986), with the additional discovery that even within the caudate itself there is fine-grained separation of multiple distinct networks that are juxtaposed and interdigitated.

Disclosures: H.L. Kosakowski: None. **N. Saadon-Grosman:** None. **R. Buckner:** None.

Poster

PSTR154. Basal Ganglia: Physiology and Function

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR154.11/DD24

Topic: E.03. Basal Ganglia

Support: NIH R37 NS040894
NIH UH3 NS103468
NIH UH3 NS129898

Title: Local evoked potentials in the subthalamic nucleus and globus pallidus during deep brain stimulation

Authors: S. SCHMIDT, J. DALE, J. J. PETERS, D. TURNER, ***W. GRILL**;
Duke Univ., Durham, NC

Abstract: Deep brain stimulation (DBS) of the subthalamic nucleus (STN) or globus pallidus internus (GPi) treats the motor symptoms of Parkinson's disease (PD). The parameters of DBS are typically adjusted by a clinician and then delivered continuously, invariant to the state of the patient, for example, changes in physical activity or medication. Closed-loop (CL-DBS) or adaptive DBS provides automatic adjustment of stimulation parameters to respond to patient state and may reduce symptoms and side effects more effectively and use less energy than continuous DBS. CL-DBS requires a feedback signal, linked to symptoms, on which to base parameter changes. Beta oscillatory activity in the local field potentials (LFPs) of the basal ganglia are correlated with symptoms of bradykinesia and rigidity and thus may serve as effective feedback signals for CL-DBS. However, beta oscillatory activity may also be modulated due to intended motor activity, change irrespective of changes in symptoms, and may even be suppressed by some symptoms, e.g., tremor. Thus, additional potential feedback signals are likely to be required to advance CL-DBS. We measured the local evoked potentials (DLEPs) in the STN and GP generated by DBS of the STN and GP. All procedures were approved by the Duke Health IRB and the FDA and the participants provided informed consent. We applied DBS individually to the STN or GPi of each hemisphere and simultaneously measured DLEPs in both nuclei with and without general anesthesia. The amplitude of the DLEP in both the GPi and in the STN decreased, and the latency of the DLEP in both nuclei increased, during 60 s of continuous high frequency GPi DBS. The responses in both nuclei were consistent with the DLEPs resulting from the reciprocal connectivity between STN and GPe (Schmidt et al. 2020). We observed greater DLEP amplitude in the right STN during GPi DBS (i.e., distal stimulation) than during right STN DBS (i.e., local stimulation). Generally we observed smaller DLEP amplitudes during the anesthetized stage two procedure, i.e., after the lead had been implanted for some weeks. These results reveal that GPi DBS produces both local DLEPs in GPi as well as in STN, and the DLEPs in STN were similar to those we characterized previously in response to STN DBS (Schmidt et al. 2020). DLEPs may have utility as a feedback signal for CL-DBS that is complementary to beta oscillatory activity. Evoked potentials appear to encode the effects of DBS on basal ganglia neurons rather than the magnitude of specific (akinetic/rigid) symptoms. Further characterization of the potential utility of DLEPs required long-term data, particularly after chronic DBS.

Disclosures: S. Schmidt: None. **J. Dale:** None. **J.J. Peters:** None. **D. Turner:** None. **W. Grill:** B. Contracted Research/Research Grant (principal investigator for a drug study, collaborator or consultant and pending and current grants). If you are a PI for a drug study, report that research relationship even if those funds come to an institution.; Boston Scientific. D. Fees for Non-CME Services Received Directly from Commercial Interest or their Agents (e.g., speakers' bureaus); Boston Scientific. E. Ownership Interest (stock, stock options, royalty, receipt of intellectual property rights/patent holder, excluding diversified mutual funds); Deep Brain Innovations, LLC.

Poster

PSTR154. Basal Ganglia: Physiology and Function

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR154.12/DD25

Topic: E.03. Basal Ganglia

Support: Saks Kavanaugh Foundation
National Institutes of Health (R01 MH060379)
William N. & Bernice E. Bumpus Foundation
Conte NIH/NIMH (P50 MH119467)

Title: Cell-type specific targeting of striato-nigro-striatal circuits using CellREADR

Authors: *A. MATSUSHIMA[1], R. WEINBERG[1], M. TRINH ORSZAG[1], E. A. HUESKE[1], N. MANGAL[1], H. SULLIVAN[1], S. E. COVEN EASTER[1], J. ALBERTA[1], X. YANG[3], I. R. WICKERSHAM[1], D. E. HOUSMAN[1], J. Z. HUANG[3], A. M. GRAYBIEL[2];
[1]MIT, Cambridge, MA; [2]MIT, CAMBRIDGE, MA; [3]Cold Spring Harbor Lab., Cold Spring Harbor, NY

Abstract: The brain is composed of numerous distinct cell-types. Individual cell-types exhibit differential morphological, electrophysiological and molecular phenotypes and interact with each other to define the function of the neural circuits in the brain. Transcriptomes play a major role in the definition and functions of the cell-types. The recent advent of CellREADR technology, which senses a specific target mRNA and then expresses an effector gene, heralded a leap in phenotype editing, enabling the targeting of specific cell-types based on their characteristic mRNA expression.
Striatum and the substantia nigra pars compacta (SNpc) form the evolutionarily highly conserved striato-nigro-striatal circuits. Key, canonical cell-types of the circuit include spiny projection neurons (SPNs) involved in dopamine D1 receptor (Drd1)-expressing direct and D2 receptor-expressing indirect pathways of the basal ganglia, SPNs residing in striosomes, the striatal compartment expressing the mu opioid receptor (Oprm1), and Aldh1a1-expressing dopamine-containing neurons in the ventral tier of SNpc (vSNpc) receiving preferential inputs from striosomal SPNs. Studies have shown that the striosomal SPNs are the sole source of direct pathway innervation of dopamine-containing neurons in vSNpc which can shut-down and evoke rebound excitation of the targeted dopamine neurons. The Aldh1a1-positive DA-containing neurons of vSNpc are the most vulnerable population in Parkinson's disease, which makes understanding their role crucial through identifying the key components of the striato-nigro-striatal circuits.
We designed CellREADR sesRNA constructs for marker genes of key striato-nigro cell types which include direct, indirect and striosomal SPNs, and vSNpc neurons: Drd1, Adora2A, Oprm1, Aldh1a1. Each construct has a different length, different number of TAG stop codons, and different relative locations of mutated stop codons. We tested 3, 2, 5, and 3 sesRNAs targeting Drd1, Adora2A, Oprm1, Aldh1a1, respectively, and verified the specificity of sesRNA

constructs as measured by the expression of GFP. For each gene, we found at least one sesRNA construct to sense the target RNA and edit the TAG stop codon in order to switch on the expression of GFP.
These results allow us to specifically record from and manipulate the canonical SPN and SNpc cell-type in vivo. CellREADR applied to specific circuits in the brain illustrates the dynamic power of this approach.

Disclosures: **A. Matsushima:** None. **R. Weinberg:** None. **M. Trinh Orszag:** None. **E.A. Hueske:** None. **N. Mangal:** None. **H. Sullivan:** None. **S.E. Coven Easter:** None. **J. Alberta:** None. **X. Yang:** None. **I.R. Wickersham:** None. **D.E. Housman:** None. **J.Z. Huang:** None. **A.M. Graybiel:** None.

Poster

PSTR154. Basal Ganglia: Physiology and Function

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR154.13/DD26

Topic: E.03. Basal Ganglia

Support: University of Michigan Parkinson Disease Research Center of Excellence
NIH R01NS109227
Brain Research Foundation Seed Grant
University of Michigan Pandemic Relief Funds

Title: Striatal dopamine dynamics regulate forelimb-digit coordination during rat skilled reaching

Authors: A. STEVENS[1], A. BOVA[1], A. TAYLOR[1], E. MUHAXHIRI[1], R. LASH[1], J. MAGNUSSON[1], C. BURGESS[2], ***D. LEVENTHAL**[1,3];
[1]Neurol., [2]Mol. and Integrative Physiol., Univ. of Michigan, Ann Arbor, MI; [3]Neurol., VA Ann Arbor Healthcare Syst., Ann Arbor, MI

Abstract: Striatal dopamine is believed to play important roles in reinforcement learning, motivation, and movement vigor (movement speed, amplitude, and frequency). However, persons with Parkinson Disease (PD) have impairments in manual dexterity that are not adequately explained by these functions. Using rat single pellet reaching to assess dexterity, we previously showed that manipulating striatal dopamine during reaches progressively alters forelimb-digit coordination, establishing a role for striatal dopamine in coordinating multi-joint goal-directed movement. With progressive nigrostriatal dopamine neuron degeneration in PD, phasic dopamine signaling is likely blunted before tonic dopamine levels decrease. Therefore, we hypothesized that dynamic dopamine signaling is critical to learn and maintain fine motor skills. To determine dynamic patterns of dopamine signaling during skilled movement, we used *in vivo* fiber photometry and a fluorescent DA biosensor (dLight1.1) to record in dorsal striatum as adult male and female rats learned a single pellet reaching task. As the rats improved at the task,

phasic dopamine release became stronger and more consistent, peaking near the time of pellet grasping. Surprisingly, there was a sharp decrease in dopamine as the forelimb retracted. Furthermore, there was a subtle difference in dopamine signaling on successful vs failed trials, with a smaller decline on successful trials. To determine the functional importance of these dynamics, we performed pilot studies during which nigrostriatal neurons were stimulated either before or after reaching. The presence or absence of dopamine neuron stimulation in a brief post-reach time window predicted subsequent reach kinematics. We conclude that dorsal striatal dopamine plays a critical role in motor adaptation more than acute motor performance, with immediate post-movement dopamine signaling causing adaptation of kinematics on subsequent trials.

Disclosures: **A. Stevens:** None. **A. Bova:** None. **A. Taylor:** None. **E. Muhaxhiri:** None. **R. Lash:** None. **J. Magnusson:** None. **C. Burgess:** None. **D. Leventhal:** None.

Poster

PSTR154. Basal Ganglia: Physiology and Function

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR154.14/DD27

Topic: E.03. Basal Ganglia

Support: NIH Grant 5R01MH125835-03
Center for Systems Neuroscience BU Individual Pstodoctoral Fellowship

Title: Context-dependent modulation of balanced population activity in striatal subtypes during visually guided locomotion in a virtual environment

Authors: *B. C. FEAREY[1], Y. TONG[1], A. S. ALEXANDER[2], Y. DING[1], **M. HOWE**[1]; [1]Psychological and Brain Sci., [2]Ctr. for Systems Neurosci., Boston Univ., Boston, MA

Abstract: As animals locomote in an environment, they continually adjust their velocity relative to changing visual cues and goals. When traversing a learned trajectory towards a goal in a familiar environment, animals initially accelerate when the goal is distant and decelerate as they approach the goal location. The striatum, the principal input nucleus of the basal ganglia, is known to modulate movement velocity and receives projections from cortical and subcortical areas, which represent the visual and motivational context. Thus, it is ideally positioned to facilitate sensori-motor transformations for appropriate context-dependent modulation of locomotion during goal pursuit. Using 2-photon calcium imaging, we measured activity simultaneously from striatal projection neurons of the direct and indirect pathways (dSPNs and iSPNs respectively) in the dorsal striatum as head-fixed mice ran on a virtual linear track to a reward location. Previous studies have described highly similar activity in dSPNs and iSPNs at locomotion onsets and offsets, arguing for complementary roles for these cell-types in promoting action. Consistent with these studies, we observed that activity in both populations increased at movement onsets at the beginning of the track and decreased at movement offsets at the end of

the track. However, the relative balance of activity in the two populations varied strongly by the movement phase and track position: dSPN activity exceeded iSPN at locomotion onsets at the track start and iSPN activity exceeded dSPN activity at offsets at the end. A proportion of SPNs of both types were reliably active at particular track locations and tiled the entire track. This location selective activity was disrupted in a different context, confirming that they are dependent on visual input. In a familiar environment, dSPN fields were more heavily weighted to the beginning of the track and iSPN fields to the end. Moreover, the movement phase specific imbalances of dSPN and iSPN were not strongly present during spontaneous initiations and terminations of movement in darkness. These data suggest that location selective imbalances in dSPN and iSPN signaling may facilitate adaptive context-specific modulation of ongoing locomotor velocity during visually guided goal-pursuit.

Disclosures: **B.C. Fearey:** None. **Y. Tong:** None. **A.S. Alexander:** None. **Y. Ding:** None. **M. Howe:** None.

Poster

PSTR154. Basal Ganglia: Physiology and Function

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR154.15/DD28

Topic: E.03. Basal Ganglia

Support: NIH R01NS094950
NSF IOS-1845355
Rutgers Busch Biomedical Grant

Title: Stable auditory-motor integration on posterior Striatum SPNs

Authors: *C. GARCÍA[1], D. J. MARGOLIS[2], **I. LINARES**[1], L. PASTERNAK[2], A. J. YONK[3]; [1]Rutgers Univ. Behavioral and Systems Neurosci., Highland Park, NJ; [2]Rutgers, New Brunswick, NJ; [3]Cell Biol. and Neurosci., Rutgers Univ. Grad. Program In Neurosci., Somerset, NJ

Abstract: A critical role of the central nervous system is to select appropriate actions based on incoming sensory information. Little is known about how our brains learn to turn meaningless auditory frequencies into action-related cues that are critical for survival. The mechanisms that allow animals to adapt to a changing auditory-outcome environment are poorly understood.The tail of the striatum (TS) is a vital area that integrates signals from the auditory cortex and thalamus and is thought to be involved in the transformation of sensation to action. The striatum contains two distinct spiny projection neuron (SPN) subpopulations (direct pathway, dSPNs and indirect pathway, iSPNs) that play essential roles in action selection and sensory value decoding. However, it is unknown how the activity dSPN and iSPN subpopulations coordinate to promote sensorimotor learning.To unveil the mysteries of these questions we designed a novel auditory-discrimination paradigm in a head-fixed setting DAAS (Dynamic Auditory Action Selection),

that allows mice to learn two different frequencies harmonics and associate them with two different actions by pushing or pulling a joystick. We recorded, with calcium imaging and two-photon microscopy, the activity of dSPN and iSPN.Our preliminary data show that animals can successfully learn the task, and medium spiny neurons encode selective auditory-action associations. SPNs show trial by trial and specific activity related to the sounds and the action. Incorrect associations and white noise don't reflect SPN associated activity. The population of SPNs are consistent to differentiate specific sound action associations. Sound-action associations are lost during reversal recordings. There is Trial by Trial consistency for the de Sound-action associations during the Kmeans of longitudinal recordings. Longitudinal cell profiles can be followed during different days, but better methodology is needed to corroborate. There is higher similarity for kmeans clusters over days than for the same neurons over days.

Disclosures: C. García: None. **D.J. Margolis:** None. **I. Linares:** None. **L. Pasternak:** None. **A.J. Yonk:** None.

Poster

PSTR154. Basal Ganglia: Physiology and Function

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR154.16/EE1

Topic: E.03. Basal Ganglia

Support: Cerebral Palsy Alliance Research Foundation Inc. (PG02518)

Title: Identifying the effective targets for deep brain stimulation: System identification using a simple recurrent neural network

Authors: *M. KASIRI[1], M. ASADI[2], T. SANGER[1,3];
[2]Dept. of electrical engineering and computer science, [1]Univ. of California, Irvine, Irvine, CA;
[3]Dept. of Neurol., Children's Health, Orange County, Orange, CA

Abstract: Deep brain stimulation (DBS) is an established neuromodulation technique for treating movement disorders. The selection of optimal targets is essential for effective outcomes and improvement of symptoms. Despite DBS's primary effect is blocking or distorting abnormal signals at the stimulation site, recent evidence shows that it also generates evoked potentials (EP) across connected brain regions, suggesting that it modulates brain functional connectivity by affecting distant targets. While numerous cortical models of human primates exist, models representing deep brain region networks are rare. Developing models of deep brain network can enhance our understanding of brain dynamics and enables us to simulate the effect of stimulation across the entire network. Recurrent neural networks (RNNs) can map local field potential signals to latent variables, revealing network dynamics. In single hidden layer networks, the weights act as dynamics matrices for state-space models, allowing for the computation of state variables and using conventional system identification techniques. In this study, we collected intrinsic brain signals (local field potential, LFPs) of basal ganglia and thalamus during voluntary

reaching, from children with dystonia, who were undergoing DBS surgery for clinical purposes. We employed an RNN model to decode network dynamics for each patient. We then used the weights of the trained models to build a state-space dynamic model of the deep brain network. The properties of this model (modes and eigenvalues) describe the stability of the network activity, and we hypothesize that unstable dynamics are a significant contributor to dystonia. We therefore used a feedforward controller to model the effect of DBS pulses on the network dynamics. We computed the likelihood that each stimulation setting would contribute to stabilizing or destabilizing the network. In other words, we used the weights derived from a nonlinear approximation method (RNN) to build a linear time invariant (LTI) system representing the deep brain network and utilized this linear model to evaluate the impact of each DBS target on the system's output. We compare the predictions of the model with actual clinical outcomes for the most effective DBS target. Our results show that network inputs that contribute to stabilizing the network dynamics are more likely to be those that lead to clinical benefit in the children with dystonia. Our approach demonstrates a valuable and simple tool for predicting optimal DBS targets in children with dystonia and holds promise for extension to other neurological conditions.

Disclosures: **M. Kasiri:** None. **M. Asadi:** None. **T. Sanger:** None.

Poster

PSTR154. Basal Ganglia: Physiology and Function

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR154.17/EE2

Topic: E.03. Basal Ganglia

Support: RFFR Grant 20-015-00482

Title: Modulation of neural activity in the external globus pallidus nucleus alters excessive oscillations in the basal ganglia-cortical motor circuits and modifies motor function in hemiparkinsonian rats.

Authors: *N. NOVIKOV[1], E. BRAZHNIK[1], I. MYSIN[1], L. SHUBINA[1], L. POPOVA[2]; [1]Inst. of Theoretical and Exptl. Biophysics, Russian Acad. of Sci., Pushchino, Russian Federation; [2]Belozersky Inst. of Physico-Chemical Biol., Moscow, Russian Federation

Abstract: Excessive synchronization of local field potential activity (LFP) in the 30-Hz range in the basal ganglia (BG) of Parkinson's disease (PD) patients, and in rodent models of PD is thought to contribute to motor dysfunction in PD. The mechanisms underlying the abnormal LFP patterns are not fully understood. Although exaggerated oscillations have been observed in several BG nuclei in PD, it is still unclear whether it manifests in the external globus pallidus (GPe), as the key nucleus of the BG network. The aim of this study was to determine the role of GPe in generating and transmitting of excessive oscillations through motor neural circuits in rat model of PD. The GPe receives inputs from the major BG nuclei, dorsal striatum (dSTR) and

subthalamic nucleus (STN) and provides back inhibitory outputs to STN, dSTR, and substantia nigra pars reticulata (SNr). This study uses the behaving rats with 6-OHDA-induced unilateral dopamine cell lesion to gain insight into the potential role of the GPe in supporting expression of LFP oscillations in the motor circuits. Electrodes were implanted in the BG nuclei, motor (MCx) and prefrontal (PFC) cortex. Cannula was inserted into the GPe in a separate group of rats to allow modulation of GPe activity via local infusion of muscimol, a GABA-A agonist, picrotoxin, a GABA-A antagonist, or glutamate receptors antagonists (GluR-ant). It was suggested that activity in the GPe contributes to the expression of exaggerated beta oscillations in the BG and may play a role in supporting PD motor symptoms. Our recent observations have shown that unlike the MCx, SNr and STN, beta oscillations are not very much evident in recordings from the GPe and dStr in PD rats, showing low coherence with recordings from MCx. Apart from beta oscillations, marked increase of coherent 50-56 Hz gamma oscillations was observed exclusively in the dStr, GPe and PFC. Also, we explored the effects of reversal modulation of neuronal activity in the GPe with local infusion of muscimol, picrotoxin, or GluR-ant on the severity of aberrant oscillations and the ability to restore locomotion in rats with PD. Noticeably, all treatments, causing inhibition or activation of neural activity in GPe, lead to strong reduction in power of beta and gamma oscillations and restoration of locomotion. In rats treated with high doses of levodopa, triggering levodopa-induced dyskinesia (LID), the infusion of muscimol to GPe abolished 100-Hz gamma oscillations in BG-cortical circuits and significantly reduced the incidence of LID. The results revealed the ultimate role of GPe, as a critical component of the BG, in controlling motor circuits activity and motor function in PD.

Disclosures: **N. Novikov:** A. Employment/Salary (full or part-time); Institute of Theoretical and Experimental Biophysics, Russia. Other; Grand from RFFR. **E. Brazhnik:** None. **I. Mysin:** None. **L. Shubina:** None. **L. Popova:** None.

Poster

PSTR154. Basal Ganglia: Physiology and Function

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR154.18/EE3

Topic: E.03. Basal Ganglia

Support: NIH Grant NS100824

Title: Neuronal dynamics of pedunculopontine GABAergic neurons during action sequence initiation and execution

Authors: *S. ZHANG, Y. KIM, D. YILMAZ, N. K. GUT, J. MENA-SEGOVIA;
Ctr. for Mol. and Behavioral Neurosci., Rutgers Univ., Newark, NJ

Abstract: The pedunculopontine nucleus (PPN) is a midbrain structure that is part of the mesencephalic locomotor region. Emerging data suggest the involvement of PPN neurons in the modulation of goal-directed behavior and the consolidation of stimulus-outcome associations.

These functions may be attributed to its bidirectional connectivity with multiple basal ganglia nuclei, including the dopamine neurons of the substantia nigra. We have recently reported that PPN_{GABA} neurons inhibit dopamine neurons and modulate goal-directed responses of unsigned valence, affecting the completion of tasks that entail both appetitive and aversive outcomes without interfering with the overall motor output. However, it is not clear whether PPN_{GABA} neurons signal the termination of a goal-directed action sequence (i.e., facilitating a switch in behavior when contingencies change), or whether they participate in shaping the execution of goal-directed behavior (i.e., tracking the progression of an action sequence). Here, we explored the group dynamics of PPN_{GABA} neurons during different stages of goal-directed behavior using fiber photometry. We transduced PPN_{GABA} neurons with a calcium indicator (GCamp) in VGat-Cre mice and we trained them in a lever-pressing task. Photometry recordings were obtained following the completion of a fixed ratio (FR) paradigm with progressive increases of the sequence length (i.e., FR1, FR3 and FR5). In the following stage of the experiment, mice were tested in a progressive ratio paradigm to record the activity of PPN neurons around the breaking point (i.e., termination of the sequence determined by the mice). We observed that PPN_{GABA} neurons increased their activity at the start of every lever-pressing sequence (FR1, FR3, FR5 and PR). Following the initial peak of fluorescence, corresponding to the first lever press, the activity decreased but remained higher than the baseline during the entire sequence. During the head entry to the reward port, the activity of PPN_{GABA} neurons increased again before returning to baseline levels. Unpurposive movement was not correlated with changes in calcium signaling. Our results support the theory that PPN_{GABA} neurons track the entire action sequence and possibly contribute to its initiation and termination. Further experiments will test the possibility of functionally different subtypes of PPN_{GABA} neurons based on their projection targets.

Disclosures: S. Zhang: None. **Y. Kim:** None. **D. Yilmaz:** None. **N.K. Gut:** None. **J. Mena-Segovia:** None.

Poster

PSTR154. Basal Ganglia: Physiology and Function

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR154.19/EE4

Topic: E.03. Basal Ganglia

Support: New Jersey Department of Health Commission for Spinal Cord Research (CSCR20IRG008)

Title: Characterization of a Glutamatergic Projection from the Pedunculopontine Nucleus to the Spinal Cord

Authors: *G. DIMARCO[1,2], N. K. GUT[2], A. SRIDHARAN[2], A. UPADHYAY[3], V. ABRAIRA[3], J. MENA-SEGOVIA[2];
[1]Rutgers Univ. Newark Aidekman Res. Ctr., Newark, NJ; [2]Ctr. for Mol. and Behavioral

Neurosci., Rutgers, The State Univ. of New Jersey, Newark, NJ; [3]W.M. Keck Ctr. for Collaborative Neurosci., Rutgers, The State Univ. of New Jersey, Piscataway, NJ

Abstract: The pedunculopontine nucleus (PPN), one of the main components of the mesencephalic locomotor region, is situated in a key location to act as an interface between upper brain structures (e.g., basal ganglia) and the spinal cord. While the PPN is neurochemically heterogeneous, its glutamatergic neurons make up the majority of PPN neurons. Recent optogenetic studies have attempted to determine the role of PPN glutamatergic (PPN_{GLU}) neuron activity in locomotion, but these studies report conflicting motor results. We have recently shown that PPN_{GLU} neurons innervate all levels of the spinal cord and may play a key role in regulating locomotion. Here, using retrograde viral tracing strategies in VGluT2-Cre mice, we found that spinal cord-projecting PPN_{GLU} neurons constitute a separate population from the PPN_{GLU} neurons that innervate basal ganglia structures. Additionally, we labeled PPN_{GLU} neurons with a Cre-dependent virus that fluorescently labels synaptophysin in pre-synaptic axon terminals and characterized the synaptic density across spinal cord regions. We found preferential innervation of the medial and intermediate zones of the upper cervical spinal cord. Further, using immunohistochemistry to identify the post-synaptic targets, we observed that PPN_{GLU} neurons target both Pax2- (inhibitory) and Lmx1b- (excitatory) positive spinal cord interneurons, but avoid contacting ChAT-positive motor neurons. In addition, PPN_{GLU} neurons also synapse onto calcium binding protein-expressing interneurons. The connectivity of the PPN-spinal cord pathway suggests that PPN_{GLU} neurons may play a role in movement preparation by regulating motor neuron excitability via the recruitment of subsets of spinal interneurons. Furthermore, our anatomical findings provide clues to explain the heterogeneous effects of activating functionally distinct subsets of PPN_{GLU} neurons, as recently reported in literature. Notably, the PPN is a target for deep brain stimulation in Parkinson's disease patients with gait impairment. Thus, understanding the organization and function of the PPN-spinal cord pathway will contribute to the assessment of novel therapeutic methods.

Disclosures: G. Dimarco: None. **N.K. Gut:** None. **A. Sridharan:** None. **A. Upadhyay:** None. **V. Abraira:** None. **J. Mena-Segovia:** None.

Poster

PSTR154. Basal Ganglia: Physiology and Function

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR154.20/EE5

Topic: E.03. Basal Ganglia

Support: NIH Grant NS100824

Title: Innervation of thalamostriatal and thalamocortical systems by cholinergic neurons of the pedunculopontine nucleus

Authors: *Y. KIM[1,2], J. MENA-SEGOVIA[1];
[1]Ctr. for Mol. and Behavioral Neurosci., [2]Behavioral and Neural Sci. Grad. Program, Rutgers University-Newark, Newark, NJ

Abstract: Cholinergic neurons of pedunculopontine nucleus (PPN) have traditionally been proposed to modulate movement and wakefulness. However, these early theories have been revised in light of recent cell-type-specific optogenetics and chemogenetics experiments, raising new questions about the function of PPN cholinergic neurons. Based on axonal density, the most prominent target of PPN cholinergic neurons is the thalamus, where virtually all thalamic nuclei receive PPN innervation. Given the recent evidence of direct PPN cholinergic innervation to the striatum, one possibility is that PPN cholinergic neurons target thalamic neurons that in turn project to the striatum (i.e., thalamostriatal, TS). Alternatively, and based on early studies of the PPN function and the regulation of brain states, the innervation by cholinergic neurons may be targeting thalamic neurons that in turn project to the cortex (i.e., thalamocortical, TC). To determine whether PPN cholinergic neurons have a preferential innervation over TC or TS systems, we designed an anatomical tracing study in ChAT-Cre rats. To label PPN cholinergic axons, we transduced cholinergic neurons with a reporter expressing the YFP. To label TS neurons, we injected the retrograde tracer FastBlue across large areas of the dorsolateral striatum. To label TC neurons, we injected a retrograde AAV transducing the reporter tdTomato and the retrograde tracer cholera toxin beta in two distinct cortical areas. Cortical areas were selected according to the preferential projection of those thalamic nuclei that receive the densest PPN cholinergic innervation, based on our recent quantification, including frontal associative, prelimbic, primary motor, secondary motor, primary sensory, and insular cortices. Quantification of axonal appositions between YFP-labeled PPN cholinergic axons and TC/TS neurons revealed that approximately a quarter of retrogradely labeled TS neurons received PPN cholinergic innervation, compared to about 13% of retrogradely labeled TC neurons. In the parafascicular nucleus (Pf), where TS neurons are primarily located, PPN cholinergic axons formed appositions with more than one third of TS neurons, and roughly one fifth of TC neurons. In the posterior thalamic nucleus, where TC neurons are more abundant, the proportion of appositions between PPN cholinergic axonal varicosities and TS and TC neurons was similar to the proportion in the Pf (one third and one fifth, respectively). Our results so far suggest that PPN cholinergic neurons preferentially innervate TS neurons over TC neurons. These results further support the modulatory role of PPN cholinergic neurons over striatal circuits.

Disclosures: Y. Kim: None. **J. Mena-Segovia:** None.

Poster

PSTR154. Basal Ganglia: Physiology and Function

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR154.21/EE6

Topic: E.03. Basal Ganglia

Support: NIH grants R01AA027768
NIH grants U01AA025932

Title: Encoding of alcohol memory in direct-pathway engram cells: A striatal cell-type-specific mechanism

Authors: *X. XIE[1], T. TAN[1], R. CHEN[3], A. CRUZ[4], W. WANG[7], H. GANGAL[5], J. LU[2], R. J. SMITH[6], L. N. SMITH[8], J. WANG[9];
[1]Texas A&M Hlth. Sci. Ctr., Bryan, TX; [2]Texas A&M Hlth. Sci. Ctr., Brayn, TX; [3]Texas A&M Univ. Syst. Hlth. Scien Neurosci. and Exptl. Therapeut., Texas A&M Univ. Syst. Hlth. Scien Neurosci. and Exptl. Therapeut., Bryan, TX; [5]Texas A&M Inst. for Neurosci., [6]Texas A&M Univ., [4]Texas A&M Univ., College Station, TX; [7]Texas A&M Univ., Baylor Col. of Med., Houston, TX; [8]Texas A&M Univ. Hlth. Sci. Ctr., Texas A&M Univ. Hlth. Sci. Ctr., Bryan, TX; [9]Texas A&M Univ. Hlth. Sci. Ctr., Texas A&M Hlth. Univ. Sci. Ctr., Bryan, TX

Abstract: The treatment to reduce relapse in alcohol abuse are limited due to an incomplete understanding of the cellular mechanisms behind long-lasting drug memories. Engram cells encode memories and contain persistent learning-induced changes. We focus on striatal direct pathway medium spiny neurons (dMSNs) in the dorsal striatum, known to mediate alcohol-seeking behaviors. Our study aims to address three questions: 1) Do direct-pathway engram cells mediate alcohol memory; 2) How does alcohol alter the recruitment and function of these engram cells; 3) How does extinction training modify relapse by impacting direct-pathway engram cells. First, we trained ArcTRAP mice to drink alcohol and tagged dMSNs activated during alcohol drinking with hM4Di agents. Chemogenetic inhibition of tagged dMSNs suppressed subsequent home-cage and operant alcohol drinking, and reinstatement of alcohol seeking. This indicates that drinking-recruited dMSNs are required for alcohol memory. Intracranial self-induction of corticostriatal long-term potentiation (LTP) promoted conditioned place preference and reinforced operant conditioning. Cue-induced reinstatement of seeking behavior further demonstrated the lasting memory formed by this LTP, suggesting that direct-pathway engrams are sufficient to encode reward memories. We then trained ArcTRAP mice for alcohol self-administration and found that alcohol drinking recruited a larger proportion of dMSNs into the engram compared to water drinking. Electrophysiological recordings revealed that alcohol intake selectively potentiated corticostriatal transmission from the medial prefrontal cortex to the drinking-recruited dMSNs. This suggests that alcohol enhances the recruitment and synaptic function of direct-pathway engram cells. In D1-Cre rats infused with Cre-dependent GCaMP7f, operant alcohol self-administration was trained. Rats underwent abstinence or extinction training, followed by a reinstatement test. Abstinence increased dMSN activity during reinstatement, which was normalized by extinction training. We also induced corticostriatal LTP after extinction training or LTD after abstinence. Post-extinction LTP and post-abstinence LTD enhanced and suppressed reinstatement of alcohol-seeking, respectively. This suggests that extinction likely silences direct-pathway engrams to suppress relapse. In summary, our findings suggest that dMSN engrams are selectively recruited by alcohol-associated behaviors and contribute to relapse. However, they can be suppressed by extinction training, reducing the relapse. This study was supported by the NIH grants R01AA027768 and U01AA025932.

Disclosures: **X. Xie:** None. **T. Tan:** None. **R. Chen:** None. **A. Cruz:** None. **W. Wang:** None. **H. Gangal:** None. **J. Lu:** None. **R.J. Smith:** None. **L.N. Smith:** None. **J. Wang:** None.

Poster

PSTR154. Basal Ganglia: Physiology and Function

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR154.22/EE7

Topic: E.03. Basal Ganglia

Support: NIH Grant R01AA027768
NIH Grant U01AA025932

Title: Optogenetic inhibition of light-captured alcohol-taking striatal engrams facilitates extinction and suppresses reinstatement

Authors: *V. VIERKANT[1], X. XIE[1], Z. HUANG[1], X. WANG[1], R. SRINIVASAN[1], Y. ZHAO[2], J. WANG[1];
[1]Texas A&M Univ. Syst. Hlth. Scien Neurosci. and Exptl. Therapeut., College Station, TX;
[2]Dept. of Translational Med., Texas A&M Univ. Syst. Hlth. Sci. Ctr., College Station, TX

Abstract: Alcohol Use Disorder (AUD) is a complex condition characterized by excessive alcohol-seeking and consumption. However, it remains unclear whether these behaviors are mediated by the same or different neural ensembles, or how one ensemble may influence the other. In this study, we used FLiCRE (Fast Light- and Calcium-Regulated Expression), a cutting-edge technique that captures neuronal ensembles activated by brief behavioral actions like lever pressing or magazine entry during operant-self administration (OSA) of alcohol. Our goal was to assess the neural substrates that underlie alcohol-seeking (lever press) and alcohol-taking (magazine entry) behaviors. First, we infused the adeno-associated viruses containing FLiCRE into the dorsomedial striatum and bilaterally implanted optical fibers in the same area of rats. This enabled us to capture and subsequently inhibit ensemble neurons optogenetically. Upon magazine entry, we delivered blue light to tag the associated neuronal ensembles. Following a three-week period post-tagging, which allowed the expression of an inhibitory opsin (eNpHR) in the tagged neurons, yellow light was delivered to inhibit their activity. We found that inhibiting OSA-tagged alcohol-taking neurons decreased both alcohol-seeking and -taking behaviors in later OSA trials. In addition, this optogenetic inhibition facilitated the extinction of alcohol-seeking behaviors. Interestingly, inhibiting these OSA-tagged alcohol-taking neurons suppressed the reinstatement of alcohol-seeking behaviors, while alcohol-taking behaviors during reinstatement remained unaffected. In summary, our findings suggest the crucial role of alcohol-taking neurons in future alcohol-seeking behaviors during both the extinction and reinstatement phases. These insights pave the way for devising novel therapeutic strategies aimed at enhancing extinction and reducing relapse to alcohol use. This study was supported by the NIH grants R01AA027768 and U01AA025932.

Disclosures: **V. Vierkant:** None. **X. Xie:** None. **Z. Huang:** None. **X. Wang:** None. **R. Srinivasan:** None. **Y. Zhao:** None. **J. Wang:** None.

Poster

PSTR154. Basal Ganglia: Physiology and Function

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR154.23/EE8

Topic: E.03. Basal Ganglia

Support: NIH Grant R01AA027768
NIH Grant U01AA025932

Title: Prenatal alcohol exposure reduces cholinergic interneuron activity and impairs cognitive flexibility in adult offspring

Authors: *W. PURVINES[1], H. GANGAL[1], X. XIE[1], A. BINETTE[1], X. WANG[2], R. MIRANDA[1], J. WANG[1];
[1]Texas A&M Univ. Neurosci. Inst. For Neurosci., College Station, TX; [2]Neurosci. and Exptl. Therapeut., Texas A&M Univ. Hlth. Sci. Ctr., Bryan, TX

Abstract: Fetal Alcohol Spectrum Disorder (FASD), caused by prenatal alcohol exposure (PAE), is associated with cognitive impairments, particularly decreased cognitive flexibility. Cholinergic interneurons (CINs) in the dorsomedial striatum (DMS) are vital for cognitive behavioral flexibility. However, the impacts of PAE on DMS CIN function and on cognitive flexibility in instrumental learning remain unexplored. In this study, we first compared the number of CINs in the DMS between PAE and control mouse offspring. We utilized ChAT-Cre; Ai14 mice, wherein ChAT-positive CINs were labeled by a tdTomato reporter. Pregnant dams underwent an intermittent two-bottle choice procedure, resulting in prenatal alcohol exposure in their offspring. Another group, receiving only water, served as a control. All offspring, aged 8 months, were perfused, and their brain sections were imaged via confocal microscopy. We found fewer tdTomato-positive CINs in the posterior DMS of PAE mice than in controls. Similar reductions in CINs were observed in both the DMS and the dorsolateral striatum using ChAT staining. To examine functional alterations, we performed patch-clamp recording and found a reduction in spontaneous CIN firing in PAE mice. These results imply that PAE reduces CIN functionality. We then investigated whether PAE affected inflexible, compulsive drinking, which might be affected by CIN activity. The PAE mice were found to consume more quinine-alcohol solution and showed a greater preference for it. Lastly, we tested whether PAE impacted the reversal of instrumental learning. Both PAE and control mice underwent two-reward conditioning: pressing the left lever for food (A1-to-O1) and the right lever for sucrose (A2-to-O2). Devaluation tests found that both groups were sensitive to outcome-devaluation, suggesting that PAE did not affect initial instrumental conditioning. However, when the contingency between actions (lever pressing) and outcomes (reward delivery) was reversed (A1-to-O2 and A2-to-O1), PAE mice were unable to learn the new contingency, as evidenced by their failure in the outcome-devaluation test, while the controls were successful. These results indicate that PAE impairs reversal learning, a behavior requiring DMS CINs, whose activity was found to be impaired in PAE offspring. Overall, our findings suggest that PAE diminishes cholinergic

function, potentially contributing to reduced cognitive flexibility in both compulsive drinking and reversal of instrumental learning. These findings advance our understanding of the pivotal role of CINs in addressing cognitive flexibility deficits in FASD treatment.

Disclosures: **W. Purvines:** None. **H. Gangal:** None. **X. Xie:** None. **A. Binette:** None. **X. Wang:** None. **R. Miranda:** None. **J. Wang:** None.

Poster

PSTR154. Basal Ganglia: Physiology and Function

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR154.24/EE9

Topic: E.03. Basal Ganglia

Support: NIH Grant R01AA027768
NIH Grant U01AA025932

Title: Bidirectional modulation of striatal cholinergic activity by direct-pathway neurons

Authors: *R. CHEN[1,2], H. GANGAL[1], X. XIE[1], R. SRINIVASAN[1], J. JONES[3], X. WANG[1], J. WANG[1,2];
[1]Texas A&M Univ. Syst. Hlth. Scien Neurosci. and Exptl. Therapeut., Bryan, TX;
[2]Interdisciplinary Fac. of Toxicology, Texas A&M Univ., College station, TX; [3]Texas A&M Univ., Dept. of Biology,, College Station, TX

Abstract: Acetylcholine (ACh), released by striatal cholinergic interneurons (CINs), plays a crucial role in mediating cognitive flexibility. Previous in vivo studies have identified a unique firing pattern of striatal CINs post-extensive instrumental conditioning, characterized by a pause followed by a rebound. GABAergic inputs, as noted in ex vivo electrophysiological studies, can induce this pause-rebound pattern in CIN firing activity. Our research aims to investigate whether GABAergic inputs from the direct-pathway medium spiny neurons (dMSNs) could elicit the observed pause-rebound pattern in ACh release. Employing slice electrophysiology and ex vivo confocal methods, we first examined the effect of dMSN stimulation on CINs. In D1-Cre; Ai32 mice, optogenetic excitation of dMSNs resulted in a pause-rebound pattern in CIN firing, with stronger stimulation eliciting a more pronounced rebound. To analyze ACh release, we infused a genetically encoded ACh sensor (rACh1.7) into the dorsomedial striatum (DMS) of D1-Cre;Ai32 mice. Confocal recordings revealed an increase in ACh releases post-optogenetic excitation of dMSNs, intensifying with stronger stimulation. These ex vivo findings suggest that GABAergic inputs from dMSNs induce a pause-rebound pattern in CIN firing activity, thereby elevating ACh release. Next, we utilized genetically encoded calcium (Flex-jGCaMP7f) and ACh (gACh4m) sensors to respectively measure dMSN activity and striatal ACh release during operant alcohol self-administration in D1-Cre rats. Our results revealed that dMSN activity increased immediately after lever presses (reward-seeking behavior), peaking within 1 second. Simultaneously, there was a transient dip in ACh activity. While some animals exhibited a

rebound in ACh activity post-dip, others did not. In summary, our findings support the notion that during instrumental training, lever presses correlate with increased dMSN activity and a dip-rebound pattern of ACh release in the striatum. This research advances our understanding of the neural mechanisms underlying cognitive flexibility and provides valuable insights into how dMSNs bidirectionally modulate ACh release during instrumental learning. This study was supported by the NIH grants R01AA027768 and U01AA025932.

Disclosures: **R. Chen:** None. **H. Gangal:** None. **X. Xie:** None. **R. Srinivasan:** None. **J. Jones:** None. **X. Wang:** None. **J. Wang:** None.

Poster

PSTR154. Basal Ganglia: Physiology and Function

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR154.25/EE10

Topic: E.03. Basal Ganglia

Support: R01AA027768
U01AA025932

Title: Distinct regulation of alcohol seeking and extinction by cortical and thalamic inputs to indirect-pathway striatal neurons

Authors: *J. WANG, T. TAN, A. BINETTE, Y. HUANG, W. PURVINES, X. XIE, R. CHEN, W. WANG, H. GANGAL, B. HENDERSON, Y. CHENG, X. WANG;
Texas A&M Hlth. Univ. Sci. Ctr., Bryan, TX

Abstract: The dorsomedial striatum (DMS), a key region implicated in alcohol use disorder (AUD), comprises medium spiny neurons (MSNs) that form the direct (dMSNs) and indirect (iMSNs) pathways of the basal ganglia. While the role of dMSNs in AUD has been extensively studied, the function of iMSNs remains largely unexplored. This study investigated the role of iMSNs in alcohol-seeking behavior and extinction learning. We found that optogenetic excitation of iMSNs decreased alcohol-seeking behavior, and long-term potentiation of inputs from the medial prefrontal cortex (mPFC) to iMSNs resulted in sustained suppression of this behavior. Moreover, extinction training strengthened glutamatergic inputs to iMSNs, while inhibiting iMSNs prevented alcohol extinction. We also identified monosynaptic inputs from the centromedian nucleus (CM) of the thalamus to iMSNs. Inhibition of CM or induction of long-term depression at CM-to-iMSN synapses impedes alcohol extinction. Our findings reveal that the mPFC-to-iMSN circuit acts as a negative regulator of alcohol-seeking behavior, while the thalamus-to-iMSN circuit plays a positive role in extinction learning. These insights into the DMS iMSN circuitry in AUD offer potential avenues for therapeutic intervention. This study was supported by the NIH grants R01AA027768 and U01AA025932.

Disclosures: **J. Wang:** None. **T. Tan:** None. **A. Binette:** None. **Y. Huang:** None. **W. Purvines:** None. **X. Xie:** None. **R. Chen:** None. **W. Wang:** None. **H. Gangal:** None. **B. Henderson:** None. **Y. Cheng:** None. **X. Wang:** None.

Poster

PSTR154. Basal Ganglia: Physiology and Function

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR154.26/EE11

Topic: E.03. Basal Ganglia

Title: A Role for the projection from the Subthalamic Nucleus to the Anterior Thalamic Nuclei in motor skill learning.

Authors: *O. OLADUNNI[1], S. ELVIG[3], S. WOLFF[2];
[1]Pharmacol., Univ. of Maryland Sch. of Med. Program in Neurosci., Glen Burnie, MD; [2]Univ. of Maryland Sch. of Med. Program in Neurosci., Baltimore, MD; [3]Univ. of Maryland Sch. of Med. Program In Neurosci., Baltimore, MD

Abstract: From serving a volleyball to typing passwords, our ability to learn and generate stereotyped movements dictates our daily interactions with our environment. Although the circuitry underlying motor skill learning and generation are not fully understood, the basal ganglia play a vital role in these processes. Within this circuitry, the Subthalamic Nucleus (STN) contributes to the suppression of unwanted movements via the indirect pathway. However, while the STN also projects to various other targets in- and outside the basal ganglia beyond the indirect pathway, the role of these projections in skill learning and execution are largely unexplored. Of particular interest is the projection to the anterior thalamic nuclei (ATN): while it has recently been implicated in the motor deficits caused by Parkinson's Disease, its role in motor skill learning and execution is entirely unexplored. To determine the physiological function of the STN-ATN projection, we took advantage of our custom motor skill learning task for rats. In this task, animals need to press a lever with a specific inter-press interval to gain a water reward. Over the course of month-long training, rats develop complex de-novo movement patterns that become spatiotemporally precise and highly stereotyped. We used a two-component viral approach to selectively silence STN neurons projecting to the ATN in naïve animals. Remarkably, silencing the STN-ATN projection dramatically sped up learning in our motor skill task. Animals reduced the variability of their developing movement patterns much faster than controls during training, allowing them to reach expert performance significantly faster. In contrast, silencing this projection in expert animals had no impact on motor skill execution. These results suggest a critical role for the STN-ATN projection in regulating kinematic variability during motor skill learning. More work will be done to determine the precise connectivity of this projection and through which pathways it influences skill learning. Further understanding of this projection and the extended motor network will not only improve our knowledge of the basic mechanisms of motor skill learning, but it may also provide insights into previously neglected circuits involved in motor disorders like Parkinson's Disease.

Disclosures: **O. Oladunni:** None. **S. Elvig:** None. **S. Wolff:** None.

Poster

PSTR154. Basal Ganglia: Physiology and Function

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR154.27/EE12

Topic: E.03. Basal Ganglia

Title: The role of a basal ganglia feedback pathway in regulating variability in motor skill learning and execution

Authors: *S. K. ELVIG, O. OLADUNNI, S. B. WOLFF;
Pharmacol., Univ. of Maryland Sch. of Med. Program In Neurosci., Baltimore, MD

Abstract: From dancing ballet to tying our shoelaces, complex motor skills are necessary for our everyday lives. The mechanisms that allow our brain to learn and execute challenging skills in a stereotyped manner remain poorly understood. The basal ganglia, a group of interconnected subcortical nuclei, are crucial for learning and executing complex motor behaviors. While most research has focused on feedforward connections within the basal ganglia, feedback connections may critically modify and refine processes involved in motor skill learning. Here we have probed an unexplored feedback connection between the Subthalamic Nucleus (STN) and the Dorsolateral Striatum (DLS). We have previously shown that precise neural activity dynamics in the DLS underlie the generation of the detailed kinematics of learned motor skills. While these representations are independent of motor cortex, feedback connections may contribute to their tight regulation, which is necessary to generate stereotypical behavior. The STN, while commonly implicated in suppressing unwanted behavior via the indirect pathway, is ideally positioned for such a role. Not only can the STN integrate information from various cortical and subcortical inputs, but it also targets the inhibitory Parvalbumin+ (PV) interneuron population in DLS. These powerful modulators can effectively constrain DLS activity dynamics. This led to our hypothesis that the STN-DLS projection provides feedback to DLS and constrains its activity to support the development and maintenance of stereotyped movement patterns. We tested this hypothesis in our rat model of motor skill learning and execution that requires the rat to press a lever two times with a specific inter-press interval (IPI) to gain a reward. Over months of training rats develop a *de novo* complex stereotyped movement pattern with rich kinematics, which allows them to reliably hit the IPI. Using a two-component retrograde viral approach to chronically and specifically silence the STN neurons that send an axon to DLS, we tested the role of this projection in our task. We found that this impaired both the acquisition and execution of the skill by increasing variability of IPIs and kinematics of the developing or previously learned complex movement patterns, respectively. To confirm this effect was due to STN's effect on DLS and not due to collateral projections, we anterogradely silenced DLS neurons that receive STN input and observed the same results. This suggests that the projection from the STN to DLS PV interneurons is necessary for developing and constraining the precise neural representations in DLS that give rise to stereotyped execution of motor skills.

Disclosures: **S.K. Elvig:** None. **O. Oladunni:** None. **S.B. Wolff:** None.

Poster

PSTR154. Basal Ganglia: Physiology and Function

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR154.28/EE13

Topic: E.01. Eye Movements

Support: NEI 1ZIA EY000415

Title: Relative value representation in the lateral part of the macaque substantia nigra pars reticulata

Authors: *A. YOSHIDA[1], O. HIKOSAKA[2];
[1]NEI, NIH, Bethesda, MD; [2]Natl. Eye Inst., Natl. Eye Inst., Bethesda, MD

Abstract: In our daily lives, we make decisions to choose or reject actions based on the values of the alternatives. However, these values can be relative, depending on the situation. For example, consider a situation where the absolute value of the three objects (Objs) to be chosen is A, B, and C, in that order (A>B>C). Two of the three Objs are presented; Objs A and B in environment X, and Objs B and C in environment Y. Obj B is relatively bad in environment X (A>B in X), but its relative value becomes good in environment Y (B>C in Y), although the absolute value remains unchanged. To investigate how such relative values are represented in the substantia nigra pars reticulata (SNr) of macaque monkeys, the output of the basal ganglia, we recorded neural activity from SNr neurons while two monkeys performed a choice task based on relative values.The task used five different Objs with different amounts of reward. In a trial, two of the five Objs are presented sequentially. At the beginning of the trial, a background picture (Environment) is presented, indicating which two Objs would be presented. When the monkeys selected an Obj, they made a saccade to the Obj, and if they fixated on the Obj for a certain duration of time (> 400 ms), they were considered to have selected the Obj, and the amount of reward was given accordingly. If they did not make a saccade to the Obj, or if they did not fixate the Obj after the saccade, they were considered to have rejected the Obj, and the next Obj was presented randomly. The next Obj was presented randomly until the monkeys finally made a choice.We found most of the task-related neurons in the lateral part of the SNr. The SNr neurons intensely decreased neural activity when the monkeys selected the relatively good Obj, while they increased neural activity when the monkeys rejected the relatively bad Obj. In other words, the SNr neurons changed their response to the same Objs in different Environments. Notably, the two monkeys often selected a relatively bad Obj in some Environments, and SNr neurons increased neural activity when they selected relatively bad Objs as well as when they rejected bad Objs. These results suggest that SNr neurons do not represent the selected behavior but rather the relative value of the presented Obj.

Disclosures: **A. Yoshida:** None. **O. Hikosaka:** None.

Poster

PSTR155. Descending and Subcortical Control and Modulation of Movement Across Behaviors

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR155.01/EE14

Topic: E.07.a. Cellular properties – Interneurons and motor neurons

Support: European Research Council (ERC) under the European Union's Horizon 2020 research and innovation programme (Grant agreement No. 95147 to D.R., A.S. and A.J.I)
Natural Sciences and Engineering Research Council of Canada (RGPIN-2017-05522 and RTI-2019-00628 to D.R.)
Fonds de la Recherche du Québec - Santé (FRQS Junior 1 awards 34920 and 36772 to D.R., FRQS Postdoctoral Training Scholarship 332188 to J.S)
Canada Foundation for Innovation (39344 to D.R.)
Centre de Recherche du Centre Hospitalier Universitaire de Sherbrooke (start-up funding and PAFI grant to D.R.)
Faculté de médecine et des sciences de la santé (start-up funding to D.R.)
Neurosciences Sherbrooke (to D.R.)
fonds Jean-Luc Mongrain de la fondation du CHUS (to D.R.)

Title: Reticulospinal fibers innervate the sublesional spinal cord in salamanders having recovered their locomotor abilities after a complete spinal cord transection

Authors: *J. SWIEGERS[1], I. KHSIME[1], C. VAN DER ZOUWEN[1], L. BLANCHÉ[1], A. JOVEN[2], A. IJSPEERT[3], A. SIMON[2], D. RYCZKO[1];
[1]Dept. of Pharmacology-Physiology, Univ. de Sherbrooke, Sherbrooke, QC, Canada; [2]Karolinska Institutet, Stockholm, Sweden; [3]École Polytechnique Fédérale de Lausanne, Lausanne, Switzerland

Abstract: Salamanders can regenerate their nervous system and recover voluntary locomotion after full spinal transection. However, the neurons carrying descending locomotor commands before and after spinal cord regeneration are unknown. Here, in the salamander Pleurodeles waltl, we studied brainstem glutamatergic reticulospinal neurons, that are known to play an important role in locomotion initiation in other vertebrates. Using tracing, in situ hybridization and immunohistochemistry, we identified reticulospinal neurons in the superior, middle and inferior reticular nuclei expressing glutamatergic markers such as glutamate, vesicular glutamate transporter 2 (Vglut2), and Chx10 (V2a neuron marker). Using whole-cell patch clamp recordings in brainstem slices, we found that most reticular neurons support tonic firing, an important characteristic of neurons mediating the descending command for locomotion. In some neurons, we imposed oscillating currents to mimic the rhythmic inputs that reticulospinal neurons receive during locomotion. The spiking activity of the recorded neurons appeared to be

entrained by the inputs at oscillating frequencies compatible with walking (0.6 -1.2 Hz) and swimming (1.3-3.3 Hz). We examined whether some glutamatergic reticulospinal neurons could control the activation of spinal motor circuits. Using calcium imaging in a brainstem-spinal cord preparation in which spinal motoneurons were labelled with calcium green, we found that stimulation of the middle reticular nucleus activated motoneurons through glutamatergic transmission. Then, we studied the in vivo locomotor role of reticular neurons using video recordings coupled with deep-learning based movement analyses. Surgical lesion of the middle reticular nucleus decreased spontaneous locomotor activity in an open field, suggesting that reticulospinal neurons in this region play an important role in locomotion initiation. Interestingly, 15 weeks after full spinal transection at low thoracic level, salamanders recovered their locomotor abilities, and such recovery was associated with the presence in the spinal cord below the lesion of reticulospinal fibers originating from the superior, middle, and inferior reticular nuclei. In the future, we will investigate whether glutamatergic reticulospinal neurons are important for such recovery in salamanders.

Disclosures: **J. Swiegers:** None. **I. Khsime:** None. **C. van der Zouwen:** None. **L. Blanché:** None. **A. Joven:** None. **A. Ijspeert:** None. **A. Simon:** None. **D. Ryczko:** None.

Poster

PSTR155. Descending and Subcortical Control and Modulation of Movement Across Behaviors

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR155.02/EE16

Topic: E.07.a. Cellular properties – Interneurons and motor neurons

Support: the European Research Council (ERC) under the European Union's Horizon 2020 research and innovation programme (grant agreement No 951477 to D.R., A.S. and A.J.I
Natural Sciences and Engineering Research Council of Canada (RGPIN-2017-05522 and RTI-2019-00628 to D.R.
Fonds de la Recherche du Québec - Santé (FRQS Junior 1 awards 34920 and 36772 to D.R.
Canada Foundation for Innovation (39344 to D.R.
the Centre de Recherche du Centre Hospitalier Universitaire de Sherbrooke (start-up funding and PAFI grant to D.R.
Faculté de médecine et des sciences de la santé (start-up funding to D.R.)
Neurosciences Sherbrooke (to D.R.)
Jean-Luc Mongrain de la fondation du CHUS (to D.R.)

Title: Restoration of locomotor movements after a full spinal cord transection in salamanders

Authors: ***I. KHSIME**[1], C. VAN DER ZOUWEN[1], A. JOVEN ARAUS[3], A. SIMON[4], A. IJSPEERT[5], D. RYCZKO[2];

[1]Dept. of Pharmacology-Physiology, [2]Univ. de Sherbrooke, Sherbrooke, QC, Canada; [3]Cell and Mol. Biol., Karolinska Inst., Solna, Sweden; [4]Dept. of Cell and Mol. Biol., Karolinska Inst., Stockholm, Sweden; [5]Ecole polytechnique federale de Lausanne, Lausanne, Switzerland

Abstract: Salamanders can swim underwater and walk on ground. They are also able to recover voluntary locomotion after full spinal transection. However, a precise description of the gradual recovery of swimming and walking movements after full spinal transection is lacking. Here we used video recordings and a deep-learning based software (DeepLabCut) to analyze the locomotor movements of the salamander *Pleurodeles waltl* before and after a full spinal transection at low thoracic level. To measure spontaneous locomotor activity, we recorded salamanders in an open field area at 30 fps. The locomotor speed and time spent in locomotion decreased after spinal transection but recovered to prelesional levels after 17 weeks. To measure locomotor movements at higher resolution, we recorded salamanders in a MotoRater at 300 fps. We tracked twenty-five anatomical points distributed on the body axis, limbs and head. To analyze axial movements, we measured the amplitude of the angular excursions along the body axis as a function of time. During swimming, a traveling wave of axial movements was propagated from head to tail. The amplitude of the angular excursions increased rostrocaudally. To establish the footfall pattern during walking, we measured the speed of limb movements. Salamanders mostly displayed a lateral sequence walk, and this was coordinated with a standing wave of axial movements in the trunk. In the first weeks after spinal cord injury, the coordination between body parts above and below the lesion was lost. After five weeks, the coordination between forelimbs and hindlimbs was restored during walking. After eight weeks, propagated waves were restored in axial body parts during swimming. Such locomotor recovery was associated with tissue regrowth at the level of the spinal cord lesion. Our work provides a detailed description of locomotor recovery in salamanders. In the future, we will couple such movement analysis with genomic, anatomical and physiological tools to understand the neural mechanisms involved in such recovery.

Disclosures: I. Khsime: None. **C. van der Zouwen:** None. **A. Joven Araus:** None. **A. Simon:** None. **A. Ijspeert:** None. **D. Ryczko:** None.

Poster

PSTR155. Descending and Subcortical Control and Modulation of Movement Across Behaviors

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR155.03/EE17

Topic: E.07.a. Cellular properties – Interneurons and motor neurons

Support: CIHR Grant 407083
NSERC Grant RGPIN-2017-05522
NSERC Grant RTI-2019-00628
FRQS Junior 1 award 34920
FRQS Junior 2 award 36772

FRQS Doctorate Scholarship BF2-335559
CFI Grant 39344
CR-CHUS startup funding and PAFI grant
FMSS startup funding
Neurosciences Sherbrooke
ERC Grant 951477

Title: Activity of glutamatergic, GABAergic and cholinergic neurons of the mammalian Mesencephalic Locomotor Region during motor behaviors and sensory events.

Authors: *C. VAN DER ZOUWEN[1], T. HSU[2], V. R. KONANUR[2], J. C. DUQUE YATE[1], M. F. ROITMAN[2], D. RYCZKO[1];
[1]Dept. of Pharmacology-Physiology, Univ. de Sherbrooke, Sherbrooke, QC, Canada; [2]Dept. of Psychology, Univ. of Illinois at Chicago, Chicago, IL

Abstract: The Mesencephalic Locomotor Region (MLR) is traditionally known to play an important role in locomotor control. However, how this region responds to sensory modalities is largely unknown. Here, we examined this question by recording the calcium activity of glutamatergic, GABAergic and cholinergic neurons in the MLR using fiber photometry in freely moving mice. In all these MLR cell types we found responses to visual stimulation (looming stimulus), auditory stimulation (unexpected brief sound) and/or mechanical stimulation (air puff). Sensory-evoked responses in MLR cells did not necessarily trigger a locomotor response. Intriguingly, during spontaneous locomotion in the open field, it was difficult to find on average a clear correlation between locomotor initiation or locomotor arrests and the mass activity of MLR neurons, although occasionally this could be seen in some animals. This suggests that single cell resolution is needed to find the correlated neurons. However, during locomotion on a motorized treadmill, we found a clear increase in calcium activity of many MLR cell types, suggesting that inputs specific to this task (possibly proprioceptive ones) drive their activity. We also found an increase in calcium activity during rearing and/or grooming in several MLR neurons. Altogether, our results suggest that the MLR receives abundant sensory inputs and comprises different sets of neurons that play a role in different aspects of motor behavior, including some behaviors beyond locomotion. Whether and how the gating of sensory information triggers such motor programs should be examined by future studies.

Disclosures: C. van der Zouwen: None. **T. Hsu:** None. **V.R. Konanur:** None. **J.C. Duque Yate:** None. **M.F. Roitman:** None. **D. Ryczko:** None.

Poster

PSTR155. Descending and Subcortical Control and Modulation of Movement Across Behaviors

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR155.04/EE18

Topic: E.07.a. Cellular properties – Interneurons and motor neurons

Support: Canadian Institutes of Health Research (407083 to D.R.)
Natural Sciences and Engineering Research Council of Canada (RGPIN-2017-05522 and RTI-2019-00628 to D.R.)
Fonds de la Recherche du Québec - Santé (FRQS Junior 1 awards 34920 and 36772 to D.R.; Doctorate Training scholarship BF2-335559 to C.I.v.d.Z.)
Canada Foundation for Innovation (39344 to D.R.)
Centre de Recherche du Centre Hospitalier Universitaire de Sherbrooke (start-up funding and PAFI grant to D.R.)
Faculté de médecine et des sciences de la santé (start-up funding to D.R.)
Neurosciences Sherbrooke (to D.R.)
European Research Council under the European Union's Horizon 2020 research and innovation program (951477)

Title: Dopamine-sensitive cell types in the mammalian Mesencephalic Locomotor Region control locomotion initiation, locomotor arrest, and turning in mice

Authors: *A. JUÁREZ TELLO, C. VAN DER ZOUWEN, L. DEJAS, J. BOUTIN, J. DUQUE YATE, J. SWIEGERS, D. RYCZKO;
Dept. of Pharmacology-Physiology, Univ. de Sherbrooke, Sherbrooke, QC, Canada

Abstract: The locomotor role of dopaminergic neurons is traditionally attributed to their ascending projections to the basal ganglia, which project to the Mesencephalic Locomotor Region (MLR), a brainstem region responsible for controlling locomotion. In addition to these ascending projections, some of us reported that descending dopaminergic projections from meso-diencephalic neurons to the MLR are present from basal vertebrates to mammals (Ryczko et al. 2013 PNAS, 2016 PNAS). In lamprey, these descending dopaminergic inputs promote locomotor output through the activation of D1 receptors in the MLR (Ryczko et al. 2013 PNAS). However, the specific neurons targeted by the descending dopaminergic projections in the mammalian MLR, as well as their behavioral role remain unknown. In this study, using virus injections, optogenetics, and deep learning-based movement analysis, we show that MLR neurons expressing D1 or D2 receptors control different aspects of movement in freely behaving mice. In the cuneiform nucleus, optogenetic activation of D1-expressing neurons promoted locomotion, whereas optogenetic activation of D2-expressing neurons stopped locomotion. In the pedunculopontine nucleus, activation of D1-expressing neurons promoted locomotion, whereas activation of D2-expressing neurons evoked ipsilateral turning. Our data uncovered in the mammalian MLR new behaviorally relevant cell types based on the expression of dopaminergic receptors. These findings likely have far-reaching implications in our understanding of how dopamine regulates movements in physiological and pathological conditions.

Disclosures: A. Juárez Tello: None. **C. van der Zouwen:** None. **L. Dejas:** None. **J. Boutin:** None. **J. Duque Yate:** None. **J. Swiegers:** None. **D. Ryczko:** None.

Poster

PSTR155. Descending and Subcortical Control and Modulation of Movement Across Behaviors

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR155.05/EE19

Topic: E.07. Rhythmic Motor Pattern Generation

Support: Lundbeck foundation R366-2021-233

Title: Neuronal population dynamics in spinal cord during PPN-induced movement arrest

Authors: *R. W. BERG[1], S. A. KOMI[2], J. KAUR[4], J. F. SØRENSEN[3];
[2]Dept. of Neurosci., [1]Univ. of Copenhagen, Copenhagen, Denmark; [3]Dept. of Neurosci., Univ. of Copenhagen, KBH N, Denmark; [4]Univ. of Copenahgen, Copenhagen, Denmark

Abstract: It was recently reported that a targeted activation of excitatory cells in the PedunculoPontine Nucleus (PPN) induces whole-body arrest of all ongoing movement in rodents. We sought to understand this mechanism and use it as a tool to control spinal motor circuits and investigate its impact on neuronal network dynamics. We sampled the activity of the lumbar spinal cord in freely moving rats using Neuropixels 1.0 and Neuronexus 128 probes. We delivered red-light stimulus to the PPN through an implanted optical fiber, where an AAV9 virus had also been injected prior, which carried a plasmid to express an opsin (ChrimsonR) under the control of the CAMKIIa promoter. Further, we identified the probe location from post-mortem FISP (Fast Imaging with Steady-state free Precession) MRI sequences and examined the spatial distribution of the population activity by delineating the local contributions of the dorsal and ventral laminae of the spinal cord. Our preliminary results indicate that during the movement arrest, there is a subset of the spinal neurons that maintain their firing as a persistent activity for as long as the stimulus occurs. The population neuronal activity exhibited rotational dynamics during locomotion, and the PPN stimulation suspended this rotation. Some neurons ceased to be active all together after the onset of PPN stimulation. Overall, the PPN-induced movement arrest offers an interesting tool for investigating spinal population dynamics.

Disclosures: R.W. Berg: None. **S.A. Komi:** None. **J. Kaur:** None. **J.F. Sørensen:** None.

Poster

PSTR155. Descending and Subcortical Control and Modulation of Movement Across Behaviors

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR155.06/EE20

Topic: E.07.a. Cellular properties – Interneurons and motor neurons

Support: DFG AC 371/2-1
DFG AC 371/1-1
Horizon 2020 MSCA-101107596

Title: In-vivo characterization of descending pathways controlling adaptive walking in Drosophila

Authors: *S. LIESSEM, S. DAHLHOFF, F. M. IQBAL, A. S. MERIC, C. J. DALLMANN, J. M. ACHE;
Neurobio. and Genetics, Theodor-Boveri-Institute, Julius Maximilian Univ. of Würzburg, Würzburg, Germany

Abstract: Adaptive locomotion is crucial for the survival of animals and is ultimately the consequence of sensorimotor processing. The circuits for generating rhythmic muscle activity patterns for locomotion are situated in the vertebrate spinal cord or the invertebrate ventral nerve cord, but the drive for initiation, modulation, and termination of movement originates in the brain. The critical link between the brain and lower-level circuits is a relatively small number of descending neurons (DNs). In *Drosophila*, inputs from different sensory modalities, including vision and mechanosensation, converge onto only ~1000 genetically identifiable DNs. Here, we take advantage of this compact, accessible system to investigate how specific neurons process multimodal sensory information to control on-going behavior. To identify pathways controlling different aspects of locomotion, we first conducted an activation and silencing screen of DNs and upstream brain interneurons (INs) in flies walking freely in an open field arena. We identified DNs and INs that control parameters that are essential for adaptive locomotion, including walking initiation and cessation, turning, and walking speed. We also found INs that elicit other, more complex behaviors such as chasing - one phase of the elaborate male courtship ritual. To understand how these and other previously identified brain neurons are recruited during behavior, we performed *in-vivo* whole cell patch clamp recordings in walking and flying flies under varying external conditions and internal states. For example, we found that DNs driving forward and backward walking are tonically active during rest and increase their activity during forward and backward walking, respectively. In both cases, spiking preceded changes in behavior. Furthermore, we found that a large portion of DNs associated with walking are gated out during flight. This suggests that walking direction (here, forward vs backward) relies on the activity of multiple DNs that are modulated by the behavioral state of the animal. In ongoing experiments, we combine physiological recordings with detailed tracking of leg kinematics to understand in detail how the DNs and INs identified in our screen drive different aspects of walking.

Disclosures: S. Liessem: None. **S. Dahlhoff:** None. **F.M. Iqbal:** None. **A.S. Meric:** None. **C.J. Dallmann:** None. **J.M. Ache:** None.

Poster

PSTR155. Descending and Subcortical Control and Modulation of Movement Across Behaviors

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR155.07/EE21

Topic: E.07.a. Cellular properties – Interneurons and motor neurons

Support: China postdoctoral science foundation（2022M722422）
Shanghai Post-doctoral Excellence Program（2022586）
Shanghai Science and Technology Innovation Program-YangFan plan（23YF1449900）
National Science and Technology Innovation 2030 Major Program (2021ZD0204500)

Title: Decoding the neural circuits of visual-motor transformation that control behavior asymmetries

Authors: *L. XU;
Tongji Univ., shanghai, China

Abstract: When running on the racetracks, visual inputs are extremely important for regulating the movement trajectory of the runners in the orbit. However how the visual information transforms into motor commands and drive the behaviors are largely unknown. Here we use larva zebrafish to study the neural mechanisms underlying vision-induced asymmetric motor behaviors. Grating drift towards the unilateral direction can reliably induce behavior asymmetries in intact animals or *ex vivo* preparations. We used unsupervised and supervised machine learning methods to accurately characterize these behaviors into three types: high angel turn (HAT), routine turn (RT) and swim. To deciphering the neural circuits controlling the turning behaviors, we used two-photon imaging and electrophysiology to explore the responsible brain regions in a large scale. A specific region in Pretectum was identified to receive the visual information from RGC and transform it into the locomotor command driving behavior asymmetry. The glutamatergic neurons in Pretectum were recruited in terms of quantity and intensity before the large angle turning behaviors were generated. To ensure the precise execution of large-angle turning, the activation of glutamatergic neurons in Pretectum excited both excitatory V2a and inhibitory glycinergic reticulospinal neurons monosynaptically. The recruitment of excitatory V2a reticulospinal neurons provides more excitation to the ipsilateral motor neurons through the ipsilateral axon projection, which contract the muscle and induce larger body bending. While the inhibitory glycinergic reticulospinal neurons were activated to suppress the activity of contralateral motor neurons via the contralateral axon projection, leading to the relaxation of contralateral body wall muscle. Thus, we revealed a neural mechanism governing the visual-motor transformation and provided a novel principle controlling behavior asymmetries.

Disclosures: L. xu: None.

Poster

PSTR155. Descending and Subcortical Control and Modulation of Movement Across Behaviors

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR155.08/EE22

Topic: E.07.a. Cellular properties – Interneurons and motor neurons

Support: Swedish Research Council 2017-02905
Wallenberg Foundation KAW 2018.0010
Swedish Brain Foundation FO2021-0317
Karolinska Institutet

Title: Functional diversity of brainstem V2a neurons in adult zebrafish

Authors: *L. MROWKA, D. MADRID-PULGARIN, L. PICTON, A. EL MANIRA; Karolinska Inst., Stockholm, Sweden

Abstract: Locomotion is a fundamental behavior relevant to many aspects of life. Studies in many model systems have unraveled the intricate organization and function of the spinal circuits controlling locomotor movements. The activity of these circuits is driven by descending neurons from the brainstem. While specific brainstem regions have been shown to control different locomotor parameters, it is not yet clear the extent to which specific brainstem neuronal populations play a dynamic role in controlling specific locomotor parameters and how they connect circuits in the spinal cord. In this study we have investigated the organization and function of brainstem V2a neurons defined by the expression of the transcription factor Chx10. Our results show that this brainstem neuronal population is highly heterogenous with diverse morphological and physiological features. Most V2a neurons that directly project to the spinal cord were located in the most caudal region of the hindbrain, while those located in the pontine region do not seem to project to the spinal cord. *In vivo* calcium imaging shows that the directly projecting neurons in the caudal hindbrain were reliably activated during spontaneous swimming and their optogenetic activation induced slow to intermediate swimming activity. In contrast, V2a neurons in the pontine region were not reliably active during spontaneous swimming and some of them were active only during unidirectional tail movements. The V2a neurons active during swimming displayed a bursting or tonic firing pattern and projected axons both caudally to the spinal cord and rostrally in the brainstem. The non-active neurons had an adapting firing with mostly local axonal projections that did not extend to the spinal cord. Our results indicate that the V2a neuronal population in the brainstem is diverse morphologically, physiologically and functionally.

Disclosures: L. Mrowka: None. **D. Madrid-Pulgarin:** None. **L. Picton:** None. **A. El Manira:** None.

Poster

PSTR155. Descending and Subcortical Control and Modulation of Movement Across Behaviors

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR155.09/EE23

Topic: E.07. Rhythmic Motor Pattern Generation

Support: NSERC RGPIN-2021-03866

Title: Systematic characterization of locomotor-related spiking activity of hindbrain V2a neurons in larval zebrafish

Authors: D. MARK, *M. KOYAMA;
Univ. of Toronto, Scarborough, Scarborough, ON, Canada

Abstract: Brain development during infancy is crucial for the establishment of complex behavior in animal species. Throughout this period, the rapidly increasing number of neuronal connections facilitates progressively intricate brain functions. Our recent research, corroborated by other studies, demonstrates that the formation of these new connections is influenced by neuronal ontogeny, leading to the sequential creation of parallel circuits. These circuits contribute unique, evolving capabilities, enabling the brain to acquire new functions in an open-ended manner without disrupting existing functionalities. Yet, how the brain dynamically orchestrates these distinct circuits for flexible behavior remains unknown. We investigate this issue by focusing on the hindbrain V2a descending neurons in larval zebrafish, using them as a model to understand circuit interaction dynamics established at varying stages of development. In this model, early-born neurons are associated with fast, powerful whole-body axial movement, while late-born neurons are involved in slower, tail-restricted axial movements. To gain insights into the precise temporal dynamics of these two circuits during their transitional phases, we have conducted systematic characterizations of their spiking activity during episodes of escape and spontaneous swimming. We achieved this through targeted loose-patch recording in Tg(vsx2:Kaede). Instead of the standard paralysis protocol involving α-bungarotoxin, we opted for the immotile mutant *relaxed* (canb1^{ts25}) to preserve the contributions of cholinergic neurons related to fast and powerful axial movements. We controlled the rate of development via temperature, ensuring that the subjects were developmentally comparable to 4 dpf at the time of recording. Our preliminary results, drawn from 44 recorded neurons primarily from the late-born population in hindbrain segments 3 to 5, reveal an unexpected heterogeneity in spiking patterns during escape and spontaneous episodes, based on their segmental position. This suggests a complex interaction between early-born and late-born populations during the transition of locomotor patterns. These insights may further our understanding of circuit organization and its role in motor behavior development.

Disclosures: D. Mark: None. **M. Koyama:** None.

Poster

PSTR155. Descending and Subcortical Control and Modulation of Movement Across Behaviors

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR155.10/EE24

Topic: E.04. Voluntary Movements

Support: R01NS123116
American Lebanese Syrian Associated Charities (ALSAC)

Title: A brain-wide map of descending inputs onto spinal V1 interneurons

Authors: P. D. CHAPMAN[1], ***A. S. KULKARNI**[1], P. DELTUVAITE[1,2], K. HAN[1], A. TREVISAN[1], L. E. FENNO[3], C. RAMKRISHNAN[4], K. DEISSEROTH[5,6,4,7], J. B. BIKOFF[1]; [1]Developmental Neurobio., St Jude Children's Res. Hosp., Memphis, TN; [2]Univ. of Bath, Bath, United Kingdom; [3]Dept. of Neurosci., Univ. of Texas at Austin, Austin, TX; [4]Dept. of Bioengineering, [5]Dept. of Psychiatry and Behavioral Sci., [6]Howard Hughes Med. Inst., Stanford Univ., Stanford, CA; [7]Wu Tsai Neurosciences Inst., Stanford Univ., Stranford, CA

Abstract: Motor output results from the coordinated activity of neural circuits in many parts of the brain, which convey information to the spinal cord via descending motor pathways. Yet our understanding of the organizational logic through which supraspinal systems target neural circuits in the spinal cord remains limited. To reveal the circuit architecture connecting descending motor systems to interneurons in the spinal cord, we performed retrograde transsynaptic tracing from V1 interneurons in the brachial spinal cord (C6-C8) using monosynaptic rabies virus. Brain-wide identification of rabies-infected neurons using serial 2-photon tomography identified over two dozen distinct brain structures providing direct innervation of V1 interneurons, including prominent input from a number of brainstem nuclei as well as cortical, cerebellar, and neuromodulatory systems. Taking advantage of our prior molecular dissection of V1 diversity in combination with intersectional viral approaches, we have begun to dissect the structure of descending inputs onto molecularly distinct subsets of V1 interneurons. These data suggest certain descending systems provide biased input to specific subsets of V1 interneurons. Collectively, our efforts shed light on the circuitry though which supraspinal pathways engage spinal motor circuits and provide a framework for future efforts aimed at dissecting functional links between the brain and spinal cord.

Disclosures: P.D. Chapman: None. **A.S. Kulkarni:** None. **P. Deltuvaite:** None. **K. Han:** None. **A. Trevisan:** None. **L.E. Fenno:** None. **C. Ramkrishnan:** None. **K. Deisseroth:** None. **J.B. Bikoff:** None.

Poster

PSTR155. Descending and Subcortical Control and Modulation of Movement Across Behaviors

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR155.11/Web Only

Topic: E.04. Voluntary Movements

Support: NIH/NIDA Grant RO1DA047637
NIH/NIA Grant R01AG050474

Title: Changes in M1 cortical parvalbumin interneuron modulation of the corticospinal tract in aged mice

Authors: *X. WEI[1], **A. GAJJAR**[2], A. PATEL[1], M. T. YAMAMOTO[1], A. SUN[2], S. HOLLEY[1], I. MODY[1], D. C. LU[1];
[1]The David Geffen Sch. of Med. at UCLA, Los Angeles, CA; [2]UCLA Chapter, Los Angeles, CA

Abstract: The primary motor cortex (M1) is composed of excitatory pyramidal (Pyr) neurons and inhibitory parvalbumin-expressing interneurons (PV-INs), which play crucial roles in local network dynamics. However, the impact of aging on the modulation of M1 PV-INs and their influence on the corticospinal tract (CST) remains uncertain. Therefore, we examined changes in PV-IN innervation in aged PV-Cre/Ai27 mice (>14 months) and their effects on CST modulation. Our findings reveal that aged mice displayed impaired motor function in balance beam and rope pulling behavior tests, indicating compromised CST pathway function. Electromyograms (eEMGs) recorded from hindlimb muscles following electrical stimulation of the hindlimb area in M1 showed that concomitant optogenetic stimulation of PV-INs enhanced eEMG activity in aged mice but had minimal effects in young mice. These results suggest a modified PV-IN modulation of CST-related activity in aging. In *ex vivo* slice recordings, light-evoked depolarizing postsynaptic potentials (PSPs) were observed in Pyr neurons only in aged mice, and were blocked by glutamate receptor antagonists APV and DNQX, but not by the GABA-A receptor antagonist gabazine. Notably, the firing patterns of light-evoked action potentials in PV-INs strongly determined the size and shape of PSPs recorded in Pyr neurons. Thus, activation of PV-INs in the aged M1 cortex facilitates CST signal transmission, suggesting an altered function of PV-INs in this region. Overall, our study provides insights into age-related neuronal circuit plasticity in the M1 network and its possible implications for motor deficits. Investigating PV-IN function may offer valuable guidance for interventions aimed at enhancing motor function in the aged.

Disclosures: X. Wei: None. **A. Gajjar:** None. **A. Patel:** None. **M.T. Yamamoto:** None. **A. Sun:** None. **S. Holley:** None. **I. Mody:** None. **D.C. Lu:** None.

Poster

PSTR155. Descending and Subcortical Control and Modulation of Movement Across Behaviors

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR155.12/EE25

Topic: E.07. Rhythmic Motor Pattern Generation

Support: NIH Grant R01NS100928
NIH Grant R01NS110550
NSERC Grant RGPIN-2016-03790

Title: Organization and operation of spinal circuits. Insights from computational modeling of tied- and split-belt treadmill locomotion in intact and spinal cord-injured cats

Authors: *I. A. RYBAK[1], N. A. SHEVTSOVA[1], J. AUDET[2], S. YASSINE[2], S. N. MARKIN[1], B. I. PRILUTSKY[3], A. FRIGON[2];
[1]Dept. of Neurobio. and Anat., Drexel Univ., Philadelphia, PA; [2]Dept. of Pharmacology-Physiology, Univ. de Sherbrooke, Sherbrooke, QC, Canada; [3]Sch. of Biol. Sci., Georgia Inst. of Technol., Atlanta, GA

Abstract: Rhythmic limb movements during locomotion are controlled by spinal central pattern generator circuits. It is considered that these circuits are composed of individual rhythm generators (RGs) for each limb, interacting through multiple commissural pathways and operating under the control of supraspinal drives and sensory feedback from the limbs. We do not fully understand the organization and operation of RGs, such as transitions between their flexion and extension states and coordination of their activity under different conditions in the intact animal and following recovery from different forms of spinal cord injury (SCI). Different competing theories exist. We constructed a computational model of spinal circuits based on known and proposed interactions between different neuron types, including genetically identified neurons from studies in mice. In our model, the RG can operate in 3 different regimes: (1) as a non-oscillating “state machine” (when external input, such as sensory feedback, is necessary to change a state, e.g., from extension to flexion), (2) as an oscillator with intrinsic rhythmicity in only one (flexor) half-center, or (3) as an oscillator of the classical half-center type, in which both flexor and extensor half-centers generate locomotor oscillations. The exact regime of each RG and coordination between them depend on the expression of intrinsic bursting mechanisms in the RG’s half-centers, commissural interactions, supraspinal drives, sensory feedback, characterizing limb loading and extending, and presynaptic inhibition from supraspinal drives controlling gains of sensory feedback. We tested our model by examining its ability to reproduce multiple experimental data on changes in the step cycle, swing and stance durations and the duty factor obtained from studies in intact, spinal-transected, and laterally hemisected cats stepping on tied- and split-belt treadmills at different speeds. The model explains multiple experimental data and provides important insights into the organization of spinal circuitry and the specific roles of central, supraspinal, and afferent interactions in the neural control of locomotion before and after different forms of SCI.

Disclosures: I.A. Rybak: None. **N.A. Shevtsova:** None. **J. Audet:** None. **S. Yassine:** None. **S.N. Markin:** None. **B.I. Prilutsky:** None. **A. Frigon:** None.

Poster

PSTR155. Descending and Subcortical Control and Modulation of Movement Across Behaviors

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR155.13/EE26

Topic: E.07. Rhythmic Motor Pattern Generation

Support: SNF Postdoc-Mobility P500PB_206824
Lundbeck foundation Grant R366-2021-233

Title: Spatio-temporal population dynamics of the lumbar spinal cord during locomotion in awake rats

Authors: *S. A. KOMI, J. KAUR, R. W. BERG;
Univ. of Copenhagen, Copenhagen, Denmark

Abstract: Mammalian locomotion relies in part on networks located in the spinal cord, which can autonomously generate rhythms and patterns. Recently, it was found that rotational neural dynamics is exhibited in ventral populations of the turtle spinal neurons controlling leg movements. However, few, if any, direct observations of large-scale spinal network dynamics in behaving mammals exist. Here, we report on the dynamics of spinal networks in rats while they engage in volitional locomotion.
We sampled the activity of the lumbar spinal cord in freely moving rats using multi-electrode arrays (Neuropixels 1.0 and Neuronexus 128 probes). It allowed us to record individual spikes in up to 450 neurons. Strikingly, the spinal network displays low-dimensional rotational dynamics, where the frequency and amplitude correlate with the locomotor output. Further, we identified the probe location from FISP (Fast Imaging with Steady-state free Precession) MRI sequences and examined the spatial distribution of the population activity by delineating the local contributions of dorsal and ventral laminae of the spinal cord to the overall dynamics. Finally, we asked whether distinct clusters of cells contributed equally to the phases of rotation
Our preliminary results suggest that the dorsal and ventral regions exhibit different forms of dynamics. Interestingly, spatial clusters demonstrate a higher phase coherence compared to electrophysiologically uniform cell groups, which displayed wider spatial distributions. In conclusion, we observed spinal rotational dynamics as a ubiquitous feature correlated with rhythmic limb movement. Moreover, our preliminary data suggest that distinct spatial areas contribute differently to sensorimotor processing.

Disclosures: S.A. Komi: None. **J. Kaur:** None. **R.W. Berg:** None.

Poster

PSTR155. Descending and Subcortical Control and Modulation of Movement Across Behaviors

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR155.14/EE27

Topic: E.04. Voluntary Movements

Support: JSPS 23H05488

Title: Neuropixels recording in the spinal cord of behaving rats

Authors: *S. EGAWA[1], A. KOSUGI[3], K. MAEDA[2], K. SEKI[4];
[1]Natl. Ctr. of Neurol. and Psychiatry, Tokyo, Japan; [2]Natl. Ctr. of Neurol. and Psychiatry, Kodaira, Tokyo, Japan; [3]NCNP, Tokyo, Japan; [4]Natl.inst.Neurosci., Tokyo, Japan

Abstract: The coordination of voluntary movements relies on an elaborated interaction between central and peripheral nervous systems, where the spinal cord assumes a critical role in modifying those neural circuits and facilitating information transmission between them. However, the precise mechanisms by which the spinal cord coordinates voluntary movements still need to be fully understood. To challenge this, we employed a high-density neural recording technique such as the Neuropixels probe to simultaneously record sensory- and motor-related neuronal activity and interneuron ones modulating them in the cervical spinal cord of rats performing arm-reaching tasks. First, we trained rats to pull a spout lever voluntarily through operant conditioning and successfully recorded a large number of neurons in the spinal cord, spanning from the dorsal to ventral horns, which was confirmed by both LFP depth profile of reactions to peripheral nerve stimulations under anesthesia and post-mortem histological analysis. Many of those neurons we recorded fired in advance, during, and/or after the movement onsets of arm-reaching tasks with phasic or tonic firings. Those were distributed biasing toward the ventral region in the spinal cord, whereas the intermediate to dorsal region showed sparse activities. Interestingly, some neurons constructed excitatory networks with hub-like functions that may contribute to modifying the motor command for controlling ongoing voluntary movements within intraspinal circuits. In future studies, we will elucidate the precise property of such neurons and their function on voluntary movements.

Disclosures: S. Egawa: None. **A. Kosugi:** None. **K. Maeda:** None. **K. Seki:** None.

Poster

PSTR155. Descending and Subcortical Control and Modulation of Movement Across Behaviors

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR155.15/EE28

Topic: E.04. Voluntary Movements

Support: Sheng Family Foundation

Title: Cerebellum supports miss-guided but not tactile-guided corrections in mouse licking

Authors: *X. HUANG, B. S. ITO, B. M. KARDON, J. H. GOLDBERG;
Neurobio. and Behavior, Cornell Univ., Ithaca, NY

Abstract: When we move our bodies, there are many types of adjustments that need to be made in real-time to achieve our goals. For example, when you reach for your phone in the dark, sometimes you miss it entirely, and must feel around where you roughly remember it to be before eventually grabbing it. Other times you might get lucky and make an initial partial contact with your pinky, which can then guide your whole hand towards the phone. Both types of

corrections are made in real-time based on sensory feedback from the contact (or lack of contact), where your hand is in space, and previous information about where the phone can be. These corrections occur on millisecond timescales, yet underlying neural mechanisms for sensorimotor control remain poorly understood. Here, we leverage advantages of the head-fixed mouse preparation to study online motor control. We combine 2-plane kHz frame rate imaging of the mouse tongue with deep learning enabled tongue segmentation to track tongue kinematics with high spatiotemporal resolution. Thirsty mice are trained to drink from a water spout, which we move at precise task moments to evoke spout misses and nicks followed by fast, adaptive reactions. Previously, our lab showed that the anterolateral motor cortex is important for miss-guided but not contact-guided corrections. Here, we focus on the cerebellum. To study miss-guided corrections, we moved the spout back after the mouse made first spout contact, before the next lick, on a random subset of trials. A trained mouse, after protruding its tongue and missing the spout at where it used to be, will produce corrective submovements (CSMs) within the same lick in order to contact the spout and get water reward. Preliminary data suggests that, when the lick-related fastigial nucleus of the cerebellum is bilaterally photoinactivated after the first spout contact, CSM probability and duration is reduced, making it less likely for mice to successfully contact the now moved spout. However, CSMs and spout contact success rate is intact in mice with bilateral fastigial lesion. To study nick-guided corrections, we moved the spout left or right instead of backwards. A trained mouse, after nicking the spout with one side of its tongue on the second lick, will re-aim its tongue on the third lick to make better contact with the spout to retrieve water. Surprisingly, preliminary data suggests that this re-aiming is intact in mice with either bilateral fastigial photoinactivated or lesioned. Together, these results suggest that real-time miss- and nick-driven corrections engage different mechanisms, and the cerebellum might be important for generating CSMs after misses, but not for re-aiming the tongue after nicks.

Disclosures: **X. Huang:** None. **B.S. Ito:** None. **B.M. Kardon:** None. **J.H. Goldberg:** None.

Poster

PSTR155. Descending and Subcortical Control and Modulation of Movement Across Behaviors

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR155.16/FF1

Topic: E.04. Voluntary Movements

Support: Jean Sheng
NSF Graduate Research Fellowship

Title: A collicular somatosensorimotor map of touch events on the tongue surface in mice

Authors: *B. ITO[1,2], Y. GAO[2], B. KARDON[2], J. H. GOLDBERG[2];
[2]Neurobio. and Behavior, [1]Cornell Univ., Ithaca, NY

Abstract: Successful actions require the use of endpoint feedback to guide future movements. While reaches can be guided by visual feedback, tongue movements are unique in that visual feedback is often unavailable, and instead require the use of tactile feedback. To examine how touch events guide tongue movements, we tracked tongue kinematics in 3D as head-fixed mice retrieved water from a moving spout. We detected the offset of tongue-spout contact on the first lick (L1) in a lick bout and randomly displaced the spout to the left or right such that the next lick (L2) would touch the spout with the left or right side of the tongue. Mice rapidly used contact location to re-aim the next lick in the bout: left contacts guided the third lick (L3) left, and right contacts guided L3 right. We used closed-loop photoinhibition to screen cortical, cerebellar, and midbrain regions implicated in directional licking. Surprisingly, bilateral inactivation of cortical and cerebellar regions during L3 did not impair touch-guided re-aiming. However, bilateral inactivation of lateral superior colliculus (lSC) halted ongoing licking, and unilateral inactivation of lSC, but not cortical regions, abolished re-aiming to the contralateral side, consistent with a role in touch-guided re-aiming. For visuomotor control, SC receives direct input from the retina and contains a retinotopic map of visual space in which stimuli in the contralateral visual field directs saccades to novel targets. Interestingly, unilateral inactivation of SC during visually-guided saccade tasks abolishes saccades to contralateral targets, similar to what we observe with lSC in our touch-guided lick task. We wondered whether lSC contains an analogous somatotopic map of touch events on the tongue surface that supports aiming licks to tactile targets. Viral tracing revealed that lSC receives input from tongue sensory neurons, and electrophysiological recordings revealed a somatotopic organization of contact events on the tongue: posterior lSC neurons were selective for contralateral contacts, whereas anterior lSC neurons were more selective for central or ipsilateral contacts, suggesting the topography of touch representations in lSC exhibit a similar organization to that known for visual stimuli. Finally, photostimulation of anterior lSC guided licks more laterally than posterior lSC, whereas photostimulation of lateral lSC guided licks more ventrally than medial lSC. Our findings demonstrate for the first time that SC contains a somatotopically-organized map of touch events on the tongue surface that is used to guide licks to tactile targets, analogous to what has been observed for visuomotor control of saccades.

Disclosures: **B. Ito:** None. **Y. Gao:** None. **B. Kardon:** None. **J.H. Goldberg:** None.

Poster

PSTR155. Descending and Subcortical Control and Modulation of Movement Across Behaviors

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR155.17/FF2

Topic: E.04. Voluntary Movements

Support: Israel Science Foundation (770/17)
NIH-BSF CRCNS (2019793)

Title: Brain-wide Characterization of Reach Behavior using fMRI and High-resolution Behavioral Monitoring

Authors: *J. ASLEH[1], A. LAWEN[2], I. KAHN[1];
[1]Zuckerman Inst., Columbia Univ., New York, NY; [2]Neurosci., Columbia Univ. Program In Neurobio. And Behavior, New York, NY

Abstract: Reaching is an everyday essential activity. Previous studies in mice have shed light on the neural mechanisms underlying reach behavior, identifying reach-associated brain regions, including the dorsal striatum. However, while having great temporal resolution, these studies look at local a priori determined regions and focused mostly on exploring stable and highly stereotyped forelimb reach behavior. Here we use functional magnetic resonance imaging (fMRI) to explore the interactions between the cerebral cortex, basal ganglia, thalamus, cerebellum, and brainstem in motor control of movement. Specifically, we focus on the dorsal striatum, comprising the caudate nucleus and putamen, which are involved in the selection and initiation of voluntary reach movements. To investigate voluntary reach, we introduce a novel experimental setup that combines whole brain fMRI with high-resolution behavioral monitoring. We employ a reach-to-water-drop behavior paradigm in mice and utilize machine learning techniques to fully characterize individual variability in reaching behavior. Our experimental design allowed us to isolate multiple components of the motor system at once, including cortical (primary and secondary motor cortices) and subcortical (e.g., striatum and thalamus). In line with existing literature, we demonstrate the involvement of these regions in voluntary reach. We identified regions within the basal ganglia, particularly the dorsal basal ganglia, that are associated with voluntary reach and primary motor cortex activation. Furthermore, we simulated a disruption in basal ganglia activity by targeting the two main neural populations of the direct and indirect pathway medium spiny neurons using chemogenetics. We measured network reconfiguration following this disruption in both animal models and control groups. Our findings underscore the importance of considering individual differences in motor performance and highlight the role of the dorsal striatum in motor decision processes. These insights contribute to our understanding of the complex motor system and provide a framework for studying motor disorders and interventions.

Disclosures: **J. Asleh:** None. **A. Lawen:** None. **I. Kahn:** None.

Poster

PSTR155. Descending and Subcortical Control and Modulation of Movement Across Behaviors

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR155.18/FF3

Topic: E.04. Voluntary Movements

Support: Craig H. Neilsen Foundation 891396
NIH 5R01NS105725

Title: Cortical sensorimotor circuits required for precise movements in mice

Authors: *S. H. TZAMOURANIS, Y. MORENO LOPEZ, E. R. HOLLIS, II;
Circuit Repair Lab., Burke Neurolog. Inst., WHITE PLAINS, NY

Abstract: Cortical sensorimotor networks integrate information from the primary somatosensory cortex (S1) with motor circuits in the primary motor cortex (M1). These intracortical networks are vital for restoring movement execution after spinal cord injury; however, current interventions for spinal cord injury often focus on restoring motor output, neglecting the critical role for somatosensory feedback in motor control. An anatomical understanding of S1-M1 corticocortical connectivity is important both for illuminating the mechanisms of healthy movement, and for revealing how spinal cord injury alters this circuitry. To elucidate the postsynaptic targets of S1 neurons in M1 layers 2/3 and 5 and determine if there are direct S1-M1 connections on corticospinal neurons, we used a dual-virus approach in combination with retrograde tracing. Using intersectional, trans-synaptic viral tracing and retrograde tracing, we found that S1 postsynaptic neurons in M1 are distributed in layers 2/3 and 5. Additionally, we identified a population of layer 5 corticospinal neurons that receive direct S1 input. These findings not only expand our understanding of sensorimotor cortical anatomy, but they suggest that S1 can directly modulate corticospinal neurons in M1. Together, these findings identify S1-M1 corticocortical circuits as promising sites for functional plasticity in response to motor learning, injury, and rehabilitation.

Disclosures: S.H. Tzamouranis: None. **Y. Moreno Lopez:** None. **E.R. Hollis:** None.

Poster

PSTR155. Descending and Subcortical Control and Modulation of Movement Across Behaviors

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR155.19/FF4

Topic: E.07. Rhythmic Motor Pattern Generation

Support: National Institutes of Health-NINDS (R01NS102920-02)
Dana & Albert R. Broccoli Charitable Foundation and Nanette and Burt Forester and Roberta Wilson

Title: Noninvasive transcutaneous spinal electrical stimulation (NTES) plus Ekso locomotor training facilitates recovery toward stepping after a severe, chronic, complex spinal injury

Authors: *K. KIJIMA[1,2], I. MONTOYA[2], H. TORRES[2], K. CHANG[2], H. ZHONG[2], R. EDGERTON[2];
[1]Dept. of Neurol., USC, Los Angeles, CA; [2]Rancho Res. Inst., Downey, CA

Abstract: Electrical transcutaneous spinal neuromodulation can generate a very wide range of physiological states of spinal and supraspinal networks at levels of stimulation well below motor

threshold. The combination of Noninvasive transcutaneous spinal electrical stimulation (NTES) and activity-based neurorehabilitation therapy (ABNT) can facilitate the reorganization of preserved neural circuits to a higher state of functional connectivity within and among multiple spinal segment levels and supraspinal networks. Given the evidence of multiple functional reorganization strategies of neural networks reported in animal models within and among spinal and supraspinal networks in facilitating motor recovery, we hypothesized that similar recovery strategies can be facilitated in a human, 10 years post-SCI (unilateral brachial avulsion plus bilaterally complete thoracic injury) when intermediately treated 2-3/week over a period of 2 years with NTES neuromodulation combined with ABNT. When performing on the NU-STEP device (bilaterally synchronized rhythmic arm-leg exercise while sitting), EMG patterns of the right limb (side of brachial avulsion) was consistently lower in amplitude than on the left side. In contrast, when stepping alternately on a treadmill or when stepping in the Ekso Bionics device the EMG amplitudes were higher on the right side. Although the amplitudes of the EMG were higher on the side of the brachial avulsion during stepping, the kinematic control of this leg was less consistent than the right. The stepping kinematics, however, became more symmetrical with training. When sitting or performing in the NU STEP which generates minimal load bearing of the lower limbs the EMG busts were more robust than the left. There was an unusual prominence of high amplitude spike potentials when at rest and during EMG bursting patterns when performing motor tasks. The frequently occurring presence of high amplitude spike potentials are also consistent with the development of aberrant connections that could contribute to poor coordination of motor pools post injury. We observed a progressively reduced incidence of high amplitude spike potentials and a more normal EMG bursting patterns with training. In summary, the combination of NTES and ABNT has the potential to facilitate the reorganization of residual corticospinal-propriospinal-spinal tracts years after injury with the likelihood of activity-dependent mechanism playing an important role in guiding newly developed networks toward higher functionality.

Disclosures: **K. Kijima:** None. **I. Montoya:** None. **H. Torres:** None. **K. Chang:** None. **H. Zhong:** None. **R. Edgerton:** E. Ownership Interest (stock, stock options, royalty, receipt of intellectual property rights/patent holder, excluding diversified mutual funds); ONWARD, SpineX.

Poster

PSTR155. Descending and Subcortical Control and Modulation of Movement Across Behaviors

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR155.20/FF5

Topic: E.07. Rhythmic Motor Pattern Generation

Support: Tsinghua-Peking Center for Life Sciences (No. 61000100123)

Title: Brainstem weaving of the whole-body 'actoms'

Authors: *J. LI[1,2,3,4], Z. ZHANG[1,5,4], J. HU[1,2,4], M. ZHOU[1,2,3,4];
[2]Sch. of Med., [3]Tsinghua-Peking Joint Ctr. for Life Sci., [4]IDG/McGovern Inst. for Brain Res., [1]Tsinghua Univ., Beijing, China; [5]Tsinghua-Peking Joint Ctr. for Life Sci., Peking Univ., Beijing, China

Abstract: Movement is eventually achieved by contractions of muscles innervated by motor neurons in the spinal cord. Each type of movement is controlled by synergetic actions of a different group of muscles at each time point, making it difficult to interpret the connections between neural codes and movements. In the brainstem, including the medulla, pons and midbrain, there are numerous motor brain regions, which have been indicated to encode diverse movements. Currently, our understanding of the structure and function of motor neural circuits in the brainstem is still very limited. We think the brainstem neural circuits play a vital role in motor control. They may receive motor decision and planning signals from upstream regions and deliver concrete movement control signals to distributed sets of motor neurons in the spinal cord. By combining 3D motion tracking, large-scale electrophysiological recordings in freely-moving mice and inverse dynamics methods to model muscle contractions, we aim to investigate how individual brainstem neurons contribute to the most basic motion components and generate diverse motor behaviors.

Disclosures: **J. Li:** None. **Z. Zhang:** None. **J. Hu:** None. **M. Zhou:** None.

Poster

PSTR155. Descending and Subcortical Control and Modulation of Movement Across Behaviors

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR155.21/FF6

Topic: B.07. Network Interactions

Support: KAKENHI 22K06475
KAKENHI 18K06528

Title: The red nucleus and its circuit network: cell identification through a target-selective approach utilizing retrograde vectors

Authors: *T. OHNO[1], H. MIYAKE[3], N. MURABE[1], S. FUKUDA[1], S. TAKAI[3], T. YOSHIDA[1], K. NAKAJIMA[1], T. HAYASHI[2];
[1]Teikyo Univ., Tokyo 173-8605, Japan; [2]Physiol., Teikyo Univ., Tokyo, Japan; [3]Dept Physiol, Teikyo Univ. Sch. Med., Tokyo, Japan

Abstract: The red nucleus consists of magnocellular neurons and parvocellular neurons, which have been thought to project to the spinal cord and inferior olive nucleus, forming the rubrospinal and rubro-olivary pathways, respectively. However, some studies have shown that both groups comprise neurons of various sizes, and more recent studies have also indicated that the rubrospinal pathway may have output from parvocellular neurons. In contrast to

magnocellular neurons and the rubrospinal pathway, parvocellular neuron and the rubro-olivary pathway has been much less studied due to the complexity of a recurrent loop involving the cerebellar nucleus. In this study, we classified the cells of the red nucleus into two groups, rubrospinal (RS) neurons and rubro-olivary (RO) neurons. We accomplished this classification by retrograde labeling of their respective targets, namely the spinal cord and inferior olive nucleus. We compared the projection and branching patterns, cell sizes, distributions within the red nucleus, and the neurotransmitter phenotypes of axons between the two cells. Initially, we retrogradely labeled RO and RS neurons using a pair of retrograde virus tracers, AAV2retro-Channelrhodopsin (ChR) 2-fluorescent proteins (GFP and RFP). These tracers were injected into the inferior olive nucleus and spinal cord, respectively. We discovered that both RO and RS neurons exhibited a range of cell size, including small and large cells. Additionally, we observed that RO neurons were widely located along the rostro-caudal axis. Secondly, we performed anterograde labeling of the axon terminals of rubro-olivary (RO) and rubrospinal (RS) neurons by injecting cholera toxin subunit b (CTB) coupled with fluorescent protein into the red nucleus. This allowed us to investigate the transmitter phenotypes associated with each axon. We employed antibodies against vesicular glutamate transporter (VGluT1 and VGluT2) and vesicular GABA transporter (VGAT) to identify excitatory and inhibitory terminals, respectively. Immunohistochemical analysis unveiled differences in the distribution within the red nucleus, projection patterns, and transmitter subtypes of RO and RS neurons.

Disclosures: **T. Ohno:** None. **H. Miyake:** None. **N. Murabe:** None. **S. Fukuda:** None. **S. Takai:** None. **T. Yoshida:** None. **K. Nakajima:** None. **T. Hayashi:** None.

Poster

PSTR156. Reaching Movement Control: Action Execution and Feedback

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR156.01/FF7

Topic: E.04. Voluntary Movements

Title: Gaze characteristics during discrete bimanual movements

Authors: *F. A. KAGERER;
Kinesiology / Neurosci. Program, Michigan State Univ., East Lansing, MI

Abstract: Research on visuomotor control of bimanual movements usually focuses on the kinematics of the hand movements; very little is known how visual attention is distributed between the hands. To study this, we had eight right-handed participants (mean age: 19.5 yrs; 6 female) and three left-handed participants (22.3 yrs; 1 female) perform simultaneous center-out reaches using two joysticks to control two cursors on a screen in front of them at a distance of 60 cm; vision of the hands was occluded. In the baseline part of the study, the cursors had to be moved from two home positions (17 cm apart) to two targets (r=0.5 mm) straight ahead (i.e., 90 deg). There were five conditions with ten trials each: both cursors visible, only right visible, only left visible, both visible with increased lateral error gain of the left-hand visual feedback, and

both visible with increased lateral error gain of the right-hand visual feedback. In the isodirectional part, the cursors had to be moved again from two home positions to two targets located at either 30 or 150 degrees; both hands moved in the same direction. In three conditions with 20 trials each, either both cursors were shown, or only right visible, or only left visible. In the anisodirectional part of the study, the target were also located at 30 and 150 degrees, but the hands had to be moved in mirror-fashion, either to the far-apart or the close-together targets; trial numbers and conditions were identical to the isodirectional part. Throughout, movement amplitude was 7.5 cm, with a 1:1 match between joystick and cursor movement. Eye movements were recorded at 120 Hz using a Tobii Pro Fusion eye tracker mounted below the monitor displaying the targets. Participants' heads were stabilized by a chin- and headrest. Each trial started with a centrally located fixation cross which disappeared when the targets came on. The variable of interest in our preliminary analysis was dwell time in the regions of interest (ROI), defined as the sum of fixations >150ms in the areas centered at the targets (ROI r=20 mm). Results show that throughout, dwell time in right-handed participants was significantly higher for the right target than for the left. The left-handed participants showed the opposite, spending more time looking at the left target. In both groups, the increased lateral error feedback shifted gaze to the more variable side. In the context of the dynamic dominance hypothesis, these results might indicate that during simultaneous discrete bimanual movements the dominant side requires more visual attention to compensate for its shortcomings in accuracy control.

Disclosures: F.A. Kagerer: None.

Poster

PSTR156. Reaching Movement Control: Action Execution and Feedback

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR156.02/FF8

Topic: E.04. Voluntary Movements

Support: Grant DG RGPIN-2019-05944
DFG FO 1347/1-1

Title: The coordination of left and right hand movements when acting on separate objects is shaped by completion for gaze.

Authors: *J. FOOKEN[1], T. ZHU[2], B. PIEKKOLA[2], J. P. GALLIVAN[3], J. FLANAGAN[3]; [1]Queen's Univ., Kinigston, ON, Canada; [2]Queen's Univ., Kingston, ON, Canada; [3]Queens Univ., Kingston, ON, Canada

Abstract: Many daily tasks, such as cooking, involve continuously using our two hands to separately grasp, move, and place various objects in target locations. In such action tasks, the objects that will be manipulated by each hand may become available (or relevant) at different times during the unfolding task. We hypothesized that the coordination of the two hands in bimanual object manipulation would be highly flexible—with each hand moving independently

as soon as a relevant object becomes available—but constrained by the competition for gaze. We designed a task in which participants used their left and right hands to separately grasp objects, located at their start locations on the left and right of midline respectively, move them straight ahead, and then drop them off by inserting them into slots. After an object was dropped off, a new object appeared at the corresponding start location following a delay. The left object delay was always 0.5 s whereas the right object delay varied, across trials, from 0.5 to 3.5 seconds. Note that with longer right object delays, the left hand was free to continue moving while the right hand awaited a new object. Each trial lasted 40 s and participants received points for each object that was dropped off. Both grasp and drop-off required precise movement, encouraging participants to fixate each object as it was grasped and each slot as an object was inserted. We found that when the left and right object delays were the same, participants moved the hands together—with a 1:1 left:right ratio—but with a temporal offset or stagger (with either the left or right hand leading). However, as the right object delay increased, the probability of the left hand moving alone increased, with the left:right ratio gradually increasing towards 2:1. In general, the temporal coordination between the hands was highly flexible and efficient, and was not constrained to a fixed ratio of 1:1 or 2:1 within a trial. Overall, the seemingly complex patterns of hand coordination, within and across trials, could be explained by a model that optimized reward and included competition for gaze resources (requiring gaze to be directed to each object as it was grasped and each slot as an object was inserted).

Disclosures: J. Fooken: None. **T. Zhu:** None. **B. Piekkola:** None. **J.P. Gallivan:** None. **J. Flanagan:** None.

Poster

PSTR156. Reaching Movement Control: Action Execution and Feedback

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR156.03/FF9

Topic: E.04. Voluntary Movements

Title: Chemogenetic activation of spinal convergent sensorimotor inputs enhances forelimb motor and somatosensory functions in monkeys

Authors: *M. SUZUKI[1], K. KOBAYASHI[3], S. I. PERLMUTTER[4], Y. NISHIMURA[2]; [2]Tokyo Metropolitan Inst. of Med. Sci., [1]Tokyo Metropolitan Inst. of Med. Sci., Tokyo, Japan; [3]Natl. Inst. for Physiological Sci., Natl. Inst. For Physiological Sci., Okazaki, Japan; [4]Univ. of Washington, Univ. of Washington, Seattle, WA

Abstract: Selective inactivation of neural activity is one of the powerful tools enabling investigation of the causal link between pathway function and behavior. In contrast, the effect on behavior of selective enhancement of pathway activity is not well-studied. Here we show chemogenetic activation of convergent inputs to the spinal cord enhances motor and somatosensory functions in macaque monkeys. We injected a retrograde viral vector expressing the excitatory designer receptors exclusively activated by designer drug (DREADD) hM3Dq

unilaterally into the cervical enlargement. Under anesthesia, deschloroclozapine (DCZ, 1 and 5 µg per kg, intramuscular injection), a highly potent and selective DREADD actuator, increased the activity of neurons in the forelimb area of primary motor cortex (M1) and of forelimb muscles in a dose-dependent manner. Furthermore, DCZ administration improved arm reaching speed and grip force in behaving monkeys. On the other hand, a higher dose of DCZ impeded dexterous finger movements such as precision grip. Moreover, the chemogenetic activation enhanced the short-latency response in forearm muscles to rapid wrist rotation, indicating that DCZ administration increased the excitability in the spinal stretch reflex circuit. In addition, DCZ administration shortened the withdrawal response latency to heat stimulation (55 ℃) applied to the monkey's hand, suggesting that the hM3Dq-mediated activation sensitized the pathway for thermal nociception. Overall results suggest that the effects of DCZ are due to the DREADD receptors being taken up by terminals of descending motor pathways, primary afferents, and motor and inter neurons in the spinal cord. These findings highlight a methodology for investigating the causal role of activity in specific neural pathways in non-human primate sensorimotor system. In addition, chemogenetic neural control opens up the possibility of a new generation of neuromodulatory therapies for restoration of impaired motor and somatosensory functions after neural damage.

Disclosures: **M. Suzuki:** None. **K. Kobayashi:** None. **S.I. Perlmutter:** None. **Y. Nishimura:** None.

Poster

PSTR156. Reaching Movement Control: Action Execution and Feedback

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR156.04/FF10

Topic: E.04. Voluntary Movements

Support: JSPS Grant-in-Aid for Early-Career Scientists #22K17756

Title: Pre-movement muscle co-contraction and motor performance deterioration under high-reward conditions

Authors: *N. SENTA[1], J. USHIBA[2], M. TAKEMI[1];
[1]Grad. Sch. of Sci. and Technol., [2]Fac. of Sci. and Technol., Keio Univ., Yokohama, Japan

Abstract: Rewards generally enhance performance by increasing motivation. However, performance can deteriorate when rewards are excessively large, as seen when a highly skilled golfer competing for a championship misses a putt of less than one meter, which they would typically not miss. The underlying motor changes in high-reward situations, where such paradoxical performance shifts occur, are not well understood. This study aimed to investigate changes in motor behavior under high-reward conditions and identify muscle activity patterns that may indicate failures in response to monetary incentives.Fourteen healthy adults participated in a motor task where the velocity of right-hand movement determined the outcomes.

Participants practiced the task for three days and were then tested on the fourth day with varying reward amounts (1, 10, 100, or 1,000 Japanese yen) offered for each trial. The frequency of the highest reward condition (1,000 yen) was set to one-seventh of the other conditions to emphasize its importance for the participants. The reward amount for each trial was informed to the participants 1-2 seconds before the beginning of each trial. Electromyographic (EMG) signals were recorded from three extensor and three flexor muscles in the right upper limb.The results showed that the success rate in the largest reward condition varied widely among participants, ranging from 20% to 80%. Thus, we divided the participants into two groups: low performers (LPs) and high performers (HPs). LPs showed higher success rates in the 100 yen condition compared to the 1,000 yen condition, whereas HPs exhibited the opposite trend. Both LPs and HPs demonstrated a significant increase in hand velocity with higher reward amounts. However, LPs displayed a significant increase in velocity variability only in the highest reward condition. Furthermore, a significant negative correlation was observed between the pre-movement co-contraction index of the biceps and triceps muscles and the success rate in the highest reward condition. This negative correlation was replicated in a subsequent experiment involving twelve new participants, suggesting that co-contraction in the upper limb muscles before movement initiation may help to predict failures caused by extremely high rewards. Future research should investigate whether biofeedback techniques that reduce muscle co-contraction before initiating movement can mitigate the paradoxical decline in motor performance associated with high rewards.

Disclosures: **N. Senta:** None. **J. Ushiba:** A. Employment/Salary (full or part-time); LIFESCAPES Inc.. E. Ownership Interest (stock, stock options, royalty, receipt of intellectual property rights/patent holder, excluding diversified mutual funds); LIFESCAPES Inc.. **M. Takemi:** None.

Poster

PSTR156. Reaching Movement Control: Action Execution and Feedback

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR156.05/FF11

Topic: E.04. Voluntary Movements

Support: PRODEX (ESA/Belspo)
Centre National d'Etudes Spatiales

Title: Movement direction drift in astronauts performing point-to-point arm movements without visual feedback

Authors: *L. OPSOMER[1,2], J.-L. THONNARD[2], P. LEFEVRE[2], J. MCINTYRE[3,4,5]; [1]Univ. catholique de Louvain, Brussels, Belgium; [2]ICTEAM and Inst. of Neuroscience, Univ. catholique de Louvain, Louvain-la-Neuve, Belgium; [3]CNRS, Paris, France; [4]Tecnalia, Basque Res. and Technol. Alliance, San Sebastian, Spain; [5]Ikerbasque Sci. Fndn., Bilbao, Spain

Abstract: Gravity provides a ubiquitous reference axis defining up and down, sensed through vestibular and somatosensory inputs. Previous studies have suggested that such an external gravitational reference helps multisensory integration by facilitating cross-modal transformations. Consistent with this hypothesis, when the head is tilted relative to gravity, performance during a cross-modal task involving eye-hand coordination is impaired. Thus, there is a need to further investigate how visual, proprioceptive and gravitational cues interact in defining a frame of reference for sensorimotor control. In particular, the effect of microgravity on spatial orientation at the level of arm motor control has been largely unexplored. We investigated the accuracy of point-to-point arm movements performed by astronauts (n=11) at multiple time points before, during and after a long-duration spaceflight (>5 months). These movements were first performed with eyes open to allow memorization of the targets and were then repeated with eyes closed. To study the interaction between visual, gravitational, and proprioceptive cues, these movements were performed in a seated or supine posture and parallel or perpendicular to the body's longitudinal axis. Movement accuracy was characterized by movement direction error, the angle between the target axis and a line connecting the start and end positions of the hand, projected on the parasagittal plane. Our data revealed a striking effect of gravity on direction error in the absence of visual feedback (eyes closed). On the ground, when the head was aligned with gravity (seated posture), direction errors were limited and stable over time. In contrast, when the head was not aligned with gravity (supine posture), direction errors grew progressively across trials, with hand trajectory gradually rotating in the parasagittal plane. Interestingly, onboard the ISS (0*g*) this movement direction drift was observed in both seated and supine posture when the eyes were closed. Furthermore, movement direction drift was still observed after several months of exposure to microgravity but went back to baseline as soon as the astronauts came back to Earth's surface. Overall, our results provide strong support for the hypothesis that gravity plays a central role in defining a reference frame for motor control, and more specifically, that gravity can serve as an anchor preventing movement drift in the absence of visual feedback.

Disclosures: L. Opsomer: None. **J. Thonnard:** None. **P. Lefevre:** None. **J. McIntyre:** None.

Poster

PSTR156. Reaching Movement Control: Action Execution and Feedback

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR156.06/FF12

Topic: E.04. Voluntary Movements

Support: PRODEX (ESA)
BELSPO

Title: Effect of gravity on the directional accuracy of point-to-point arm movements

Authors: *S. VANDERGOOTEN[1,2], L. OPSOMER[2], J.-L. THONNARD[4], J. MCINTYRE[5,6], P. LEFEVRE[3];

[1]UCLouvain, Louvain-la-Neuve, Belgium; [2]ICTEAM and Inst. of Neuroscience, Univ. catholique de Louvain, Brussels, Belgium; [3]ICTEAM and Inst. of Neuroscience, Univ. catholique de Louvain, Louvain-la-Neuve, Belgium; [4]Inst. of Neuroscience, Univ. catholique de Louvain, Brussels, Belgium; [5]Ctr. Natl. de la Recherche Scientifique, CNRS, Paris, France; [6]Basque Res. and Technol. Alliance, Tecnalia, San Sebastian, Spain

Abstract: On Earth, gravity plays a primordial role during motor planning and control, both as a reference axis and as a driving force for motor optimization. To further explore how the central nervous system takes gravity into account, we studied the influence of body orientation relative to gravity on the accuracy of point-to-point arm movements to remembered target locations.
We performed three experiments. For all the experiments, participants performed point-to-point arm movements parallel to the longitudinal body axis towards visual targets, first with eyes open (for memorization of the target locations), then with eyes closed.
In the first experiment, participants were seated on a chair that could be rotated backward by 45° or 90° in order to assess the influence of body orientation. We observed that when the body was not aligned with gravity (45 and 90° conditions), errors in movement direction, i.e., the angle between the target axis and the line joining the onset and offset positions of a movement, increased gradually across trials performed with eyes closed (decreasing accuracy).
In the second experiment, participants were either seated on a chair or lying on their back in supine position and we investigated whether movement direction error increased as a function of time or as a function of movement repetition. Results showed that the error increased as a function of time, since the total increase in movement direction error was the same whether participants performed 6 or 24 movements over the course of a 45-second block.
In the third experiment, participants were lying on their back in supine position and we investigated whether performing movements aligned with body axis but without targets would also result in movement direction error. We observed in our data similar patterns of error with or without targets, suggesting that the horizontal direction itself was not perceived accurately.
Altogether, our results show an increase over time in movement direction error for point-to-point arm movements when the body is not oriented along the vertical axis (in the absence of visual feedback), perhaps suggesting an influence of vestibular information on maintaining a stable representation of body orientation in space.

Disclosures: S. Vandergooten: None. **L. Opsomer:** None. **J. Thonnard:** None. **J. McIntyre:** None. **P. Lefevre:** None.

Poster

PSTR156. Reaching Movement Control: Action Execution and Feedback

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR156.07/FF13

Topic: E.04. Voluntary Movements

Support: NIH Grant 2R15HD093086

Title: Three-dimensional supplemental kinesthetic feedback can improve target capture accuracy when reaching without vision

Authors: *R. N. MAZOROW, K. D. BASSINDALE, R. A. SCHEIDT;
Biomed. Engin., Marquette Univ. & Med. Col. of Wisconsin, Milwaukee, WI

Abstract: Decreased sensations of limb position and movement are experienced by ~50% of stroke survivors, leading to a decrease in upper extremity motor control and difficulty performing common daily tasks. Although visual feedback of the arm and hand can partly compensate, resulting movements are typically slow and jerky. We propose an alternate compensatory approach: using supplemental vibrotactile feedback about hand position to enhance closed-loop control of the arm. Previous work has shown that healthy individuals and some stroke survivors can use 2-dimensional (2D) feedback to improve reaching accuracy without visual feedback. Based on these results, we hypothesized that training with 3-dimensional (3D) vibrotactile feedback will lead to post-training improvements in reaching accuracy without concurrent vision compared to proprioception alone. Twelve neurologically healthy humans (7 female, 24.8±3.4 years) reached towards a visually indicated target with their dominant hand supported against gravity while holding a squeeze ball with an integrated active infrared marker. The view of the arm and hand were blocked. Reaches were performed under different feedback conditions and participants squeezed when they believed they reached the target. A motion tracker (OPTOTRAK 3020) collected real-time reach-to-grasp movements and converted them to cursor motions on a vertical display. The screen showed a 5x5 grid of targets (±x, ±y) with each location having 5 concentric rings representing the depth (±z) requirement for the reach. Six eccentric rotating mass motors were placed on the stationary non-dominant arm; each motor produced a vibrational signal representing cursor deviations from the center of the workspace along one direction (±x, ±y, ±z). Three hours of training took place over three sessions spanning three consecutive days. We tested this hypothesis via a-priori t-tests between baseline and post-training measurements. Vibrotactile feedback was effective in promoting increased accuracy of goal-directed reaches in the absence of concurrent visual feedback of hand motion. Post-training vibrotactile feedback reduced total Euclidean target capture error compared to the baseline vibrotactile block ($p = 0.008$) and the post-training proprioception block ($p < 0.001$). These results demonstrate the utility of 3D supplemental vibrotactile kinesthetic feedback and help to establish a training timeline for healthy participants. By characterizing the improvement of healthy participants over time, we will be able to optimize protocols for post-stroke training.

Disclosures: R.N. Mazorow: None. **K.D. Bassindale:** None. **R.A. Scheidt:** None.

Poster

PSTR156. Reaching Movement Control: Action Execution and Feedback

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR156.08/FF14

Topic: E.04. Voluntary Movements

Title: Impact of visual-proprioceptive perception in virtual reality on visuomotor integration during overhand throwing: a pilot study

Authors: M. CHENG, *L. CHUKOSKIE;
Northeastern Univ., Boston, MA

Abstract: Autistic adolescents engage less in physical activity and more in sedentary behavior than neurotypical peers, causing increased risks of obesity and cardio-metabolic health concerns. Visuomotor integration (VMI) challenges, manifesting as motor coordination difficulties, are evident from infancy in autistic individuals and can further last a lifetime, which may limit participation in common physical activities such as ball sports. Autistic individuals exhibit superior local visual sensitivity but demonstrate inefficient, bottom-up global visual information processing that relies heavily on visual input rather than prior knowledge, causing incongruent VMI during tasks that require precise timing such as motion perception and action sequence perception. Exergames in virtual reality (VR) have the potential to promote physical activities in autistic adolescents. However, little is known about how autistic individuals' visual perception and visual information processing in VR impact VMI during gross motor tasks. We first used a spatial estimation task to assess the visual-proprioceptive accuracy in a static VR environment. We then used an overhand ball-throwing task, a commonly performed physical activity in both the physical environment (PE) and in exergames, with eye and movement tracking to compare the VMI process during movement planning and execution in VR versus an equivalent PE. Additionally, the study explored the use of a visually-simplified VR environment to assess if reducing visual clutter could improve VMI during overhand throwing. In a pilot study with eight non-autistic young adults, we observed intact proprioceptive accuracy but reduced visual and visual-proprioceptive accuracy in VR compared to PE. Throwing success rates were lower in VR, with more gaze scanning of the environment and shorter fixation duration on task-relevant objects. Participants spent a longer time preparing for movement execution and had higher hand velocity at ball release in VR. Reducing visual clutter did not improve throwing accuracy but reduced scanning of the environment and increased fixation duration on task-relevant objects during movement planning and execution phases. These pilot results identified the differences in visual-proprioceptive accuracy and VMI between VR and PE in non-autistic adults and supported further exploration of the potential benefits of reducing visual clutter in visual information processing during overhand throwing. Subsequent research will explore the impact of autistic traits and visual-proprioceptive perception on VMI performance in VR gross motor tasks.

Disclosures: M. Cheng: None. **L. Chukoskie:** None.

Poster

PSTR156. Reaching Movement Control: Action Execution and Feedback

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR156.09/FF15

Topic: E.04. Voluntary Movements

Support: H2020-EIC-FETPROACT-2019-951910-MAIA

Title: Efficient motor learning in tool use tasks through deep kinematic inference

Authors: *M. PRIORELLI, I. P. STOIANOV;
Inst. of Cognitive Sci. and Technologies, Natl. Res. Council, Padua, Italy

Abstract: In normal conditions, the kinematic plant of an agent remains constant or only gradually changes on a lifetime scale. However, it could also change in fast timescales, for example, when using a tool to solve a task. When monkeys are trained to use a tool to reach an object, their internal bodily representations in parietal and motor areas change to represent the tool. It is thus critical to understand how the motor cortex can take into account and predict such rapid changes in the kinematic plant, and to efficiently simulate the same scenario in robotic experiments. In Optimal Control theories, complicated cost functions usually have to be defined to tackle such dynamic elements, but it seems unlikely that the same mechanisms are at work in biological organisms. In contrast, Predictive Coding based theories such as Active Inference, which tackles the motor control inversion by generating proprioceptive predictions from high-level latent states, provide a simpler and more biologically plausible solution that does not use any cost function. The advantages of the Active Inference framework are evident when using hierarchical models, which are able to construct a more complex representation of the environment, allowing the agent to better predict the observations and act over the world. Nonetheless, the current literature lacks implementations of hierarchical models, in particular regarding kinematic structures for realistic settings. We propose that, if a hierarchical model is designed where each block encodes information about a segment of the kinematic plant, the agent is capable of realizing complex trajectories in an efficient way, inferring the correct kinematic configuration during perception, and modifying its kinematic plant online by adding or removing joints at specific locations of the kinematic hierarchy. The simultaneous learning of the joint angles and limb lengths during the movement is made possible through cycles of perception and action, allowing the agent not to get stuck during the free energy minimization process, which happens when both phases are not run rhythmically. Such architecture may be required for an agent to solve a multi-step task involving reaching a tool, grabbing it (e.g., extending the length of the last joint), and finally reaching an object with its extremity. Standing on a solid theoretical basis, our novel model provides interesting hypotheses about motor control, tool use, and the role of cortical oscillations.

Disclosures: M. Priorelli: None. **I.P. Stoianov:** None.

Poster

PSTR156. Reaching Movement Control: Action Execution and Feedback

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR156.10/FF16

Topic: E.04. Voluntary Movements

Support: NIH NINDS Grant R01NS053606

Title: Isometric reaching for use in post stroke spasticity therapy

Authors: *A. RAMIREZ[1], N. AGHAMOHAMMADI[2], A. CANCRINI[1], C. CELIAN[2], B. BORGHI[1], A. MELENDEZ-CALDERON[4], J. L. PATTON[3];
[1]Univ. of Illinois at Chicago, Chicago, IL; [2]Shirley Ryan AbilityLab, Chicago, IL; [3]bioengineering (UIC); Ctr. for neuroplasticity (SRALab), Shirley Ryan AbilityLab, Winnetka, IL; [4]Univ. of Queensland, Brisbane, Australia

Abstract: Isometric Reaching for Use in Post Stroke Spasticity Therapy
Arturo Ramirez[12], Naveed Reza Aghamohammadi[12], Bruno Borghi[12], Adriana Cancrini[12],Courtney Celian[2], Alejandro Melendez-Calderon[3], James L. Patton[12]
[1] University of Illinois at Chicago, Chicago, Illinois 60607[2] Shirley Ryan AbilityLab, Chicago, Illinois 60611 [3] University of Queensland, Brisbane, Australia
Post-stroke rehabilitation should maximize motion range, function, and independence, and it has been shown that patients benefit from repetitive physical or mental training. [1,2] Patients with post-stroke spasticity respond particularly well to force modulated training and constraint-induced therapy to address learned non-use due to the spasms inhibiting dose recommended movement in repetitive physical training.[1,2]One advantage is visual feedback of movement without actual movement, possibly allowing individuals with spasticity to train without spasms. We propose using a robotic simulation of arm dynamics to represent the appropriate movements using a virtual avatar. Recent evidence has shown the potential of visual feedback to create the sensation of limb movement (despite being still), leading to adaptation.[4,5] By leveraging this phenomenon, this framework can increase functional recovery in spastic patients, helping them efficiently learn the best internal models for arm movement. Using the *b.u.r.t.* robot (Barrett Medical, Watertown, MA) with the *Unity* game engine for visual feedback, we create a haptic spring to secure individuals' limb motions into an isometric condition, then have them repetitively move the virtual hand to displayed targets. Measured forces drive an arm dynamic simulation producing movements of an avatar arm on a first-person augmented reality screen mounted over their physical arm. We hypothesize that (1) this approach allows movements not otherwise possible given the limitations of spasticity, and (2) we expect to see an improvement in real-life actions once the person's limb is released. The ability to change from constrained to unconstrained movement any time affords an opportunity to assess the impact of sensory deception using isometric conditions. This framework has the potential to develop novel treatments and provide better insights on the mechanisms of spasticity. **References:**1. Francisco GE, McGuire JR, (2012) Stroke 43(11), 3132-3136.2. Butler AJ, Page SJ (2006) APMR 87(12), 2-11.3. Kagawa S et al. (2013) Journal of stroke and cerebrovascular diseases, 22(4): 364-70.4. Bourdin P, Martini M, Sanchez-Vives MV, Scientific Reports 9(1), 1-9.5. Melendez-Calderon A, Bittmann M, Patton JL, (2017) 118 (1), 219-233.

Disclosures: A. Ramirez: None. **N. Aghamohammadi:** None. **A. Cancrini:** None. **C. Celian:** None. **B. Borghi:** None. **A. Melendez-Calderon:** None. **J.L. Patton:** None.

Poster

PSTR156. Reaching Movement Control: Action Execution and Feedback

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR156.11/FF17

Topic: E.04. Voluntary Movements

Title: The role of spinal excitability on sense of force production.

Authors: *B. H. KOPICKO[1], C. K. THOMPSON[1], W. G. WRIGHT[1], I. KURTZER[2]; [1]Hlth. and Rehabil. Sci., Temple Univ., Philadelphia, PA; [2]New York Inst. of Technol. - Col. of Osteo. Med., Old Westbury, NY

Abstract: Voluntary motor output is influenced by both central motor drive and spinal motoneuron excitability. The sense of force production has been purported to arise from corollary discharges of motor drive. If motoneuron excitability significantly contributed to the sense of force production, higher levels of excitability would require less motor drive, yielding underestimates of force output. We therefore sought to assess the perceptual consequences of voluntary force generation during protocols where spinal excitability is likely increased. We hypothesize that voluntary force generation during increased motoneuron excitability will result in a lower perception of force production. During several experiments, time-varying force fields were applied to the upper extremity of participants using a KinArm Endpoint Lab. The task involved maintaining a movable cursor within a stationary target using a manipulandum and resisting dynamic ramps and static holds. We assessed sense of force production using several reporting methods: force matching, numeric rating, identifying force direction, verbal report of offloading, and graphical recall. Contrary to our hypothesis, our results point to an overall perceptual accuracy or overestimation of force. Thirty-one participants accurately identified the amplitude of a static matching force to a reference force, after exposure to an intervening high-force ramp (i.e. reference force > high-force ramp > static force). When asked to rate the intensity of force on a numeric scale, the reported errors significantly overestimated similar amplitude forces if preceded by a high-force ramp. They were also accurate in identifying the direction of a force as it offloaded and reversed, and verbally identified the instantaneous time when an offloading force reversed directions. Finally, when asked to graphically recall the force profile, participants drew accurate or non-significantly greater force estimates than the actual force. These results indicate overall perceptual accuracy or overestimation of forces occurring at times when spinal excitability is presumed to be augmented. Given the current finding, it is likely that compensatory factors influence the perception of force generation in the upper extremity, possibly to a greater extent than spinal excitability.

Disclosures: B.H. Kopicko: None. **C.K. Thompson:** B. Contracted Research/Research Grant (principal investigator for a drug study, collaborator or consultant and pending and current grants). If you are a PI for a drug study, report that research relationship even if those funds come to an institution.; NIH Grant: RO1NS125863. **W.G. Wright:** None. **I. Kurtzer:** None.

Poster

PSTR156. Reaching Movement Control: Action Execution and Feedback

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR156.12/FF18

Topic: E.04. Voluntary Movements

Support: NIGMS Grant P20-GM-109098
NIGMS Grant P30-GM-103503
NIGMS Training Grant T32AG052375
NIGMS Training Grant T32-GM-081741
NIGMS Grant U54-GM-104942

Title: Quantifying changes in muscle activity related to postural and propulsive forces produced during reaching by people with chronic hemiparesis

Authors: *A. S. KOROL[1], A. ADCOCK[2], V. GRITSENKO[3];
[1]Neurosci., [2]Neurol., [3]Human Performance - Physical Therapy, West Virginia Univ., Morgantown, WV

Abstract: Arm hemiparesis after a stroke reduces the quality of life by impairing arm function. This impairment is caused due to changes in the muscle activity patterns as a result of the neurological damage after stroke. Even though each stroke is unique, common patterns of abnormal muscle coactivations are observed, such as flexor synergy. Many secondary impairments develop over time like spasticity that are also the result of the abnormal coactivation patterns. In our previous work, we introduced a novel method to separate movement components describing arm support against gravity and arm propulsion during movement using biomechanics underpinning. We found that anti-gravity forces were produced by consistent groups of muscles, especially in the non-dominant limb, while the propulsive forces were produced by fewer immutable synergies. Here, we will address the question of how muscle coactivation patterns change after stroke, specifically in supporting a limb against gravity and propulsion. We use principal component analysis to extract features representing postural (anti-gravity) and propulsion components of electromyography (EMG) of twelve muscles recorded during reaching by the healthy (n=10) and stroke participants (n=8, where n=3 with left hemisphere stroke and n=5 with right hemisphere stroke). Next, we analyzed the changes in eigenvalues representing these features across multiple reaching directions and muscles. We found increased coactivation between more pairs of muscles in stroke in comparison to controls, evidenced by linear relationships between corresponding eigenvalues. The increased coactivation was present during the postural and propulsion components of EMG. These results highlight the potential as a quantitative metric of muscle function after stroke.

Disclosures: A.S. Korol: None. **A. Adcock:** None. **V. Gritsenko:** None.

Poster

PSTR156. Reaching Movement Control: Action Execution and Feedback

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR156.13/FF19

Topic: E.04. Voluntary Movements

Support: National Sciences and Engineering Research Council of Canada
Canadian Foundation for Innovation
Ontario Research Fund

Title: Intermittent robotic guidance during an auditory-cued rhythmic sequential task: Effects on spatial vs. temporal accuracy

Authors: *A. MOSTOFINEJAD, R. GOODMAN, T. LORIA, M. THAUT, L. TREMBLAY; Univ. of Toronto, Toronto, ON, Canada

Abstract: Many attempts have been made to use robotic guidance to improve sensorimotor performance. Recently, we developed a paradigm in which robotic guidance is interspersed with unassisted practice (i.e., mixed practice or intermittent guidance). Such intermittent guidance was first shown to be more effective than robotic guidance alone for the learning of a golf-putting task (Bested et al., 2019a & b). Based on our interest in upper-limb, music-based rehabilitation, we also showed that intermittent guidance can also acutely improve timing accuracy within a rhythmic, circular movement task (Mostofinejad et al., 2023). However, the latter task could be viewed as more associated with emergent timing and the previous study did not include delayed retention tests. Thus, the current study investigated the effectiveness of intermittent guidance on the learning of spatiotemporal parameters of an auditory-cued, rhythmic sequential task. That task involved spiral movement sequences made up of half-circles of different sizes. It was hypothesized that using intermittent guidance would lead to better spatial and temporal accuracy in a 24hr retention test. Participants (n = 26) were seated and held a custom-built stylus over a table. The task was to synchronize their spiral movements with rhythmic auditory tones. A robotic arm could also provide physical guidance. On day one, during the pre-test, all participants completed 8 trials of a spiral movement sequence. During the acquisition phase, participants were divided into two groups and practiced the spiral movement task for 96 trials. In the unassisted group, participants completed the acquisition trials on their own (i.e., no robotic guidance). Participants in the intermittent guidance group were assisted by the robot on 50 percent of the trials (i.e., switching after each set of 12 trials), while being asked to actively follow the robotic arm. After the acquisition, all participants completed an immediate retention test, which was identical to the pre-test. On day two, participants completed a delayed retention test and a transfer test, which involved performing a novel spiral sequence. Analyses revealed that during the delayed retention test, the intermittent guidance group had a significantly smaller spatial error as compared to the unassisted group. However, there were no significant differences between the groups in terms of temporal accuracy. In conclusion, as compared to unassisted practice, intermittent guidance seems to be more effective for learning the spatial characteristics, while being comparably effective for learning the temporal characteristics of an auditory-cued, rhythmic sequential task.

Disclosures: A. Mostofinejad: None. **R. Goodman:** None. **T. Loria:** None. **M. Thaut:** None. **L. Tremblay:** None.

Poster

PSTR156. Reaching Movement Control: Action Execution and Feedback

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR156.14/FF20

Topic: E.04. Voluntary Movements

Title: Exploring the influence of object motion and smooth pursuit eye movements on posture stabilizing mechanisms during collision between moving objects and the hand

Authors: *O. SINHA[1], S. K. COLTMAN[1], T. S. ROSENQUIST[1], J. P. KPANKPA[2], A. R. FEDORSHAK[1], I. KURTZER[3], T. SINGH[1];
[1]Kinesiology, Pennsylvania State Univ., State College, PA; [2]Kinesiology, The Pennsylvania State Univ., State College, PA; [3]Dept. of Biomed. Sci., New York Inst. of Technol., Old Westbury, NY

Abstract: During many activities of daily living, humans frequently interact with moving objects. These interactions require processing the motion of inertial objects and applying force over short periods of contact to change their state of motion. Interception studies have provided valuable insights into the link between eye movements and kinematic aspects of movements, revealing a strong role of smooth pursuit eye movements (SPEM) in guiding movement trajectories. However, it is unclear if SPEM also facilitates kinetic aspects of motor control, such as how the motor system stabilizes posture against the reactive forces generated during contact. In a previous study, we found that SPEM are important for anticipatory posture stabilization prior to contact between the hand and virtual moving objects. In the current exploratory study, we explore how SPEM might affect posture-stabilizing mechanisms during limb motion to force control transitions. Healthy adult participants grasped a robotic manipulandum with their right hand and were instructed to stop virtual objects moving at different speeds. They first initiated an interception movement towards the object and then applied a brief force pulse at contact. We measured eye movements, limb movement kinematics, muscle activity in five upper limb muscles, and reactive forces at contact. We found that participants' hand trajectories and the angle of hand-to-object contact prior to the collision changed with learning as the task progressed. Furthermore, object speed appeared to influence angle of hand-to-object contact. These results provide a framework to probe the mechanisms of how motion-to-force transition occurs during contact between moving objects and the hand.

Disclosures: O. Sinha: None. **S.K. Coltman:** None. **A.R. Fedorshak:** None. **I. Kurtzer:** None. **T. Singh:** None.

Poster

PSTR156. Reaching Movement Control: Action Execution and Feedback

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR156.15/FF21

Topic: E.04. Voluntary Movements

Support: NIH NINDS GRANT R01NS053606
SWISS National Center Of Competence in Research (NCCR))

Title: Modeling Sources of Sensory Contributions in Error-Based Learning through AR and Robotic Treatment

Authors: *A. CANCRINI[1,2], N. AGHAMOHAMMADI[2,1], A. RAMIREZ[1,2], B. BORGHI[1,2], C. CELIAN[2], J. L. PATTON[3];
[1]Univ. of Illinois at Chicago, Chicago, IL; [3]bioengineering (UIC); Ctr. for neuroplasticity (SRALab), [2]Shirley Ryan AbilityLab, Chicago, IL

Abstract: Recent advancements in medicine have highlighted the importance of identifying specific impairments in stroke survivors to improve treatment outcomes. Sensory function plays a crucial role in motor deficits and recovery potential after a stroke. For example, individuals may experience deficits in proprioception or touch; sensory reweighting allows the motor system to attend differently to different senses. Personalized therapy should begin with assessing an individual's sensory contributions to error-based learning, since personalized sensory augmentation has been shown to enhance learning and reduce errors more effectively. Developing an individualized mathematical model can not only help understand and rectify sensory deficits, it can be used to personalize training environments, improving treatment outcomes and patient recovery. We employ the he *b.u.r.t.* robot (Barrett Medical, Watertown, MA) with the *Unity* game engine with augmented reality display technology to present first-person 3D actions. Mixing physical and virtual worlds allows for selective manipulation of sensory feedback, and such distortions can differentiate the sources and relative contributions to an individual’s learning tendencies. This can lead to personalized optimal control strategies for training. This framework can explain how an individual’s sensory integration influences their learning and can greatly aid in the development of future interventions that promote learning.

Disclosures: A. Cancrini: None. **N. Aghamohammadi:** None. **A. Ramirez:** None. **B. Borghi:** None. **C. Celian:** None. **J.L. Patton:** None.

Poster

PSTR156. Reaching Movement Control: Action Execution and Feedback

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR156.16/FF22

Topic: E.04. Voluntary Movements

Support: Internal DCC grant
NWA, nr. 1292.19.298

Title: Tracking the peripheral preparatory motor state during perceptual decision making using corticomuscular coherence

Authors: *Y. F. VISSER, W. P. MEDENDORP, L. P. J. SELEN;
Donders Inst. for Brain, Cognition and Behavior, Radboud Univ., Nijmegen, Netherlands

Abstract: Decision making and motor control are two sides of the same coin. Recent work has shown that even spinal reflexes are already pre-tuned during the formation of a decision. Cortico-muscular coherence (CMC), which characterizes the functional connection between the motor cortex and muscles, has been used to quantify preparatory state of the motor periphery and has been shown to increase for the muscle relevant for performing the response. However, in these experiments responses were discrete and unambiguous while in many cases we must select an action based on a continuous stream of information. Using random dot motion (RDM) stimuli, which allows for a slow buildup of the available sensory information, it has been shown that there is a continuous flow of decision related information to central motor areas. Currently there are no data on whether the peripheral preparatory state reflected in the CMC also tracks the decision state. In this study, participants (n=40) watched an RDM pattern and were asked to indicate the perceived motion direction by moving a handle to one of two targets, requiring flexion or extension of the elbow. We took EEG and EMG measurements and computed time evolving CMC between the left motor cortex and the right arm bi- and triceps. Preliminary data analyses show that decisions about stimuli with higher dot coherences were made faster and more accurately than those with lower dot coherence. Additionally, we find that CMC builds up over time as evidence accumulates towards a decision. We hypothesize that higher dot coherences are associated with a faster build-up of CMC, explaining the increase in performance. Our findings would confirm the continuous flow hypotheses, as well as provide a new way to quantify the temporal evolution of a decision variable in the peripheral motor system.

Disclosures: Y.F. Visser: None. **W.P. Medendorp:** None. **L.P.J. Selen:** None.

Poster

PSTR156. Reaching Movement Control: Action Execution and Feedback

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR156.17/FF23

Topic: E.04. Voluntary Movements

Support: Swedish Research Council (VR Starting Grant 2019-01909)
Marie Sklodowska-Curie Individual Fellowship

Title: Action does not enhance but attenuates predicted touch

Authors: X. JOB[1], ***K. KILTENI**[2];
[1]Neurosci., Karolinska Inst., Stockholm, Sweden; [2]Donders Inst. for Brain, Cognition and Behaviour, Nijmegen, Netherlands

Abstract: Dominant motor control theories propose that the brain uses efferent information to predict and attenuate the somatosensory consequences of actions, referred to as sensory attenuation. Support for this model comes from psychophysical and neuroimaging studies showing that touch applied on a passive hand elicits attenuated perceptual and neural responses if it is generated by actively tapping with one's other hand, compared to identical touch from an external origin. However, recent experimental findings have challenged this view by providing psychophysical evidence that the perceived intensity of touch on the passive hand is enhanced if the active hand does not receive simultaneous tactile stimulation with the passive hand (somatosensory enhancement) and by further attributing attenuation effects to the double tactile stimulation of the hands upon contact. Here, we directly contrasted the hypotheses of the attenuation and enhancement models regarding how action influences somatosensory perception by manipulating whether the active hand contacts the passive hand. In three preregistered experiments, we demonstrate that action does not enhance the perceived intensity of touch (Experiment 1), that the previously reported "enhancement" effects are driven by the baseline condition used (Experiment 2), and that self-generated touch is robustly attenuated regardless of whether the two hands make contact (Experiment 3). Our results provide conclusive evidence that action does not enhance but attenuates predicted touch. These findings prompt a reappraisal of recent experimental findings upon which theoretical frameworks proposing a perceptual enhancement by action prediction are based.

Disclosures: X. Job: None. **K. Kilteni:** None.

Poster

PSTR156. Reaching Movement Control: Action Execution and Feedback

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR156.18/FF24

Topic: E.04. Voluntary Movements

Support: Canadian Institutes for Health Research grant #PJT-175063 (LS)
VISTA
York University Athletic Therapy staff and students

Title: Sex-related differences in cognitive motor integration deficits as a function of collision versus contact sport participation

Authors: C. A. MARKS[1], N. SMEHA[2], M. MODICA[2], ***L. E. SERGIO**[2];
[1]Psychology, [2]Kinesiology and Hlth. Sci., York Univ., Toronto, ON, Canada

Abstract: Simple, direct interactions with the environment are contingent on our brain processes that require intact connections between frontal, parietal, and subcortical brain regions. The

integrity of these connections is crucial during the execution of more complex movements that require cognitive-motor integration (CMI), where the guiding visual information and motor action are decoupled. Following concussion, the integrity of these networks may be compromised,[1,2] resulting in an impaired ability to integrate rules into coordinated motor tasks. Further, athletes with a history of concussion show greater alterations in functional connectivity in networks associated with sport participation.[3] Finally, sex-related differences have been observed in (a) brain networks which control CMI,[4] and (b) post-concussion recovery of visuomotor skill[5]. Here, we examine the interaction between sex, CMI, and level of sub-concussive impact exposure. We hypothesized that there would be a sex-related difference in performance as a function of sport type. Participants: 155 healthy varsity athletes (85F) who participated in either collision (n = 75) or contact (n = 80) sports. Participants performed a visuomotor task where the viewed location of the target and the finger motion were decoupled. In addition, there was a visual feedback reversal between cursor motion and finger motion. We observed that males had worse cognitive-motor integration performance relative to females, regardless of sport type, on reaction time, movement time, and accuracy ($p>0.05$). In contrast, females had significantly worse performance only in collision sports on these same variables relative to females in contact sports ($p<0.01$), such that their performance was similar to males in collision sport. These data suggest that in males, concussive/sub-concussive impact exposure may be similar between contact and collision sport, contributing to brain network dysfunction in controlling skilled performance. While the exact source of the neural control difference - whether impact frequency or impact force - remains uncertain, the resilience observed in females diminishes upon engaging in collision sports.1.Cook MJ et al. Neuroimage clinical. 2020 Jan 1;25:10212. 2.Sergio LE et al. (2020). Frontiers in Neurology. 2020 Sep 16;11:1060.3.Churchill NW et al. Front in Neurol doi:10.3389/fneur.2017.00390.4.Gorbet D & Sergio (2007). EJN 25(4), 1228-1239. 5.Pierias A et al. (2020). Cognitive-motor integration performance testing and symptom assessment reveals males and females respond differently to concussive injury. Canadian Concussion Centre 8th Annual Concussion Research Symposium.

Disclosures: C.A. Marks: None. **N. Smeha:** None. **M. Modica:** None. **L.E. Sergio:** None.

Poster

PSTR157. Connectivity

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR157.01/FF25

Topic: E.07.a. Cellular properties – Interneurons and motor neurons

Support: NIH Grant R01MH117808

Title: Comprehensive reconstruction of leg motor control circuits in Drosophila

Authors: *A. P. COOK[1], A. W. AZEVEDO[1], E. LESSER[1], L. ELABBADY[1], J. S. PHELPS[2], S. KURODA[2], B. MARK[1], A. MOUSSA[1], C. J. DALLMANN[1], S. AGRAWAL[1], S.-Y. J. LEE[1], B. PRATT[1], W.-C. A. LEE[2,3], J. C. TUTHILL[1];

[1]Dept. of Physiol. and Biophysics, Univ. of Washington, Seattle, WA; [2]Dept. of Neurobio., [3]F.M. Kirby Neurobio. Center, Boston Children's Hosp., Harvard Med. Sch., Boston, MA

Abstract: Animal movement is controlled by motor neurons (MNs), which project out of the central nervous system to activate muscles. While the overall morphology of MNs is known for many animals, the identity of the neurons that provide synaptic input to these MNs remain unknown across the animal kingdom. The small size of the *Drosophila* nervous system makes this organism ideal for premotor circuitry analysis. *Drosophila* leg and wing MNs reside within the ventral nerve cord (VNC), which is analogous to the vertebrate spinal cord. We reconstructed all 69 left front leg MNs and their presynaptic partners using an automated serial section electron microscopy (ssEM) dataset of the *Drosophila* female adult nerve cord (FANC). We completed this reconstruction by automatically segmenting neural morphology with convolutional neural networks, then correcting errors made by the automated segmentation by merging and cutting neuronal segments. Our results show that the dominant input to MNs are local interneurons (single neuromere) together with input from intersegmental interneurons , descending neurons , ascending neurons, and sensory neurons . The morphology of these premotor neurons allowed us to determine their developmental origin, and therefore the neurotransmitter they release. Premotor neurons release GABA, acetylcholine, or glutamate, which establishes both inhibitory and excitatory control of MN activity. Descending neurons provide ~10% of the input to each MN, which is constant regardless of the total amount of synaptic input an individual MN receives. We also found that leg MNs receive monosynaptic input from sensory neurons, including proprioceptors and rouch receptors. This work represents the first comprehensive analysis of a premotor connectome in any limbed animal, and lays the foundation for future studies in other limbed animals.

Disclosures: A.P. Cook: None. **A.W. Azevedo:** None. **E. Lesser:** None. **L. Elabbady:** None. **J.S. Phelps:** None. **S. Kuroda:** None. **B. Mark:** None. **A. Moussa:** None. **C.J. Dallmann:** None. **S. Agrawal:** None. **S.J. Lee:** None. **B. Pratt:** None. **W.A. Lee:** None. **J.C. Tuthill:** None.

Poster

PSTR157. Connectivity

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR157.02/GG1

Topic: E.07.a. Cellular properties – Interneurons and motor neurons

Support: Missouri Spinal Cord Injury/Disease Research Program 22-03

Title: Longitudinal structure of V2a output to spinal neurons

Authors: *S. BELLO ROJAS[1], A. BERTRAM[2], M. W. BAGNALL[3];
[1]Neurosci., Washington Univ. In St. Louis, Saint Louis, MO; [2]Neurosci., Washington Univ. in St. Louis, St. Louis, MO; [3]Washington Univ., Washington Univ., Saint Louis, MO

Abstract: In vertebrates, excitatory spinal interneurons are essential for the initiation and propagation of locomotor activity. Ipsilaterally projecting V2a (Vsx2+) neurons provide a major component of this excitatory wave. Activation of V2a neurons evokes motor activity, whereas V2a neuron ablation eliminates locomotor responses. Despite the importance of V2a neurons to locomotor network function, the specific targets and structure of connections along the rostro-caudal axis remain unknown. In this study, we used optogenetic stimulation of transgenic larval zebrafish with a digital micromirror device (DMD) to map synaptic outputs of V2a neurons onto several distinct classes of identified spinal neurons: fast and slow motor neurons, V1 inhibitory neurons, V0v excitatory neurons, and other V2a neurons. We find that V2a neurons form both short and long range connections, but the strength of connectivity varies along the length of their axons. Descending connections from V2a neurons onto both fast and slow motor neurons are weighted to longer ranges, where the peak evoked synaptic activity occurs following stimulation between 3-5 muscle segments rostral from the neuron recording site. Similarly, V2a targets receive their strongest synaptic input from other V2a neurons located 4-6 segments rostrally. Here we show for the first time that two of the preferential targets of ascending V2a excitation, which arises from a subset of bifurcating V2a neurons, are slow motor neurons and V0v (Evx1+) neurons. Additionally, we tested the prediction from computational modeling that V2a neurons excite V1 (Eng1+) neurons. We show the first physiological evidence of this connection, where V1 neurons receive their strongest synaptic input from V2a neurons located 3-4 segments rostrally. We conclude that V2a neuron connectivity varies along the longitudinal axis, with peak synaptic output occurring at 3-6 segments caudal from the V2a cell body but with systematic variations in target identity. We are currently testing additional postsynaptic targets, including inhibitory V2b (Gata3+) and dI6 (Dmrt3a+) neurons.

Disclosures: S. Bello Rojas: None. **A. Bertram:** None. **M.W. Bagnall:** None.

Poster

PSTR157. Connectivity

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR157.03/GG2

Topic: E.07.a. Cellular properties – Interneurons and motor neurons

Support: Swedish Research Council 2017-02905
Wallenberg Foundation KAW 2018.0010
Swedish Brain Foundation FO2021-0317
Karolinska Institute

Title: Origin and organization of commissural inhibition that coordinates locomotion in adult zebrafish

Authors: *P. FONTANEL, M. BERTUZZI, L. D. PICTON, A. EL MANIRA;
Neurosci., Karolinska Inst., Stockholm, Sweden

Abstract: Animals rely on flexibility to successfully navigate their environment. In adult zebrafish, speed flexibility arises from the sequential recruitment of three-speed modules (slow, intermediate, and fast) composed of motoneurons (MNs) and ipsilateral excitatory interneurons (V2a INs). While the circuit organization for speed control is well-described, mechanisms underlying locomotor coordination across and along the body are still unclear. Here, we first characterize the function of two glycinergic inhibitory commissural populations, V0d and dI6, involved in the left-right alternation. In the adult zebrafish, both populations are heterogeneous and divided into three speed-dependent subclasses. Notably, the V0d population exhibited a bias toward slower speeds, while the dI6 INs predominantly belonged to the fast module. We then mapped the connectivity pattern of both populations using optogenetic stimulation at different rostrocaudal positions in the spinal cord while performing patch-clamp recordings of MNs, V2a, V0d and dI6 INs. This approach revealed both local and long-range inhibitory connections onto the contralateral populations underlying the rhythm and pattern of locomotion. Our results show that V0d INs provide strong long-range inhibition to MNs, while they provide uniform inhibition to V2a INs independent of the position along the spinal cord. In contrast, dI6 INs provided the strongest inhibition locally to V2a INs. Finally, we mapped the organization of the premotor drive from V2a to V0d INs using dual patch-clamp recordings. We show strong and more frequent excitatory connections between interneurons belonging to the same speed module, suggesting a modular organization of the premotor excitation to V0d INs, similar to the excitation pattern of MNs. We are also assessing the ipsilateral drive from V2a to dI6 INs using a similar approach. In summary, our study thus far uncovered three distinct patterns of commissural inhibition based on the source and the target that could play an important role in the coordination of locomotion in a speed-dependent manner.

Disclosures: P. Fontanel: None. **M. Bertuzzi:** None. **L.D. Picton:** None. **A. El Manira:** None.

Poster

PSTR157. Connectivity

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR157.04/GG3

Topic: E.07.a. Cellular properties – Interneurons and motor neurons

Support: NIH Grant R00NS114194

Title: Simple structural motifs can explain many features of spinal circuit activity during locomotion

Authors: *B. LEMBERGER[1], J. MURRAY[1], D. L. MCLEAN[2];
[1]Univ. of Oregon Inst. of Neurosci., Eugene, OR; [2]Northwestern Univ., Evanston, IL

Abstract: Neural activity in the spinal cord during locomotion exhibits sequences of neural bursts traveling down the body from segment to segment coordinated with alternation of activity from the left to the right. The time scales of these two dynamics are linked via a relationship

known as “constant phase-lag”, but they are also flexible and can be co-modulated by top-down signals from the brain. Previous modeling work has successfully reproduced these essential features by making use of complex, biologically realistic model neurons with heterogeneous single-cell properties and connectivity weights. In this work, we investigate the role network structure alone can play in reproducing these features by constructing an inhibition-stabilized neural population featuring homogeneous single-neuron properties and simple structural motifs of connectivity. This model simultaneously performs rhythmogenesis, generates variable-speed segment-to-segment propagation with the proper constant phase-lag relationship, and executes left/right alternation, without the fine-tuning of single-neuron or connectivity parameters. A version of the model with separate populations of fast- and slow-swim preferring neurons can replicate the recruitment of speed modules with respect to swim frequency, and the speed of swimming can be varied via targeted drive to the fast or slow module. These results suggest the possibility that much of the phenomenology of the spinal circuit can be understood from the perspective of connectivity motifs, without necessarily relying on detailed information about heterogeneous single-cell properties.

Disclosures: **B. Lemberger:** None. **J. Murray:** None. **D.L. McLean:** None.

Poster

PSTR157. Connectivity

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR157.05/GG4

Topic: E.07.a. Cellular properties – Interneurons and motor neurons

Support: Swedish Research Council 2017-02905
Wallenberg Foundation KAW 2018.0010
Swedish Brain Foundation FO2021-0317
Karolinska Institutet
StratNeuro

Title: The role of intraspinal proprioceptor feedback for flexible locomotor rhythm-generation

Authors: L. PICTON[1], ***D. MADRID-PULGARIN**[1], A. PAZZAGLIA[2], J. PATRICK ARREGUIT O'NEILL[2], A. FERRARIO[2], A. IJSPEERT[2], A. EL MANIRA[1];
[1]Karolinska Inst., Stockholm, Sweden; [2]École Polytechnique Fédérale de Lausanne, Lausanne, Switzerland

Abstract: Proprioceptive feedback is important for the function of motor networks and allows animals to adjust motor commands based on real movement outcomes and in light of environmental cues. We previously identified a population of inhibitory, Piezo2-expressing proprioceptors located inside the spinal cord of adult zebrafish that are activated upon bending of the body. These intraspinal proprioceptors provide direct inhibitory feedback to the excitatory, rhythm-generating V2a interneurons that is essential for normal locomotion *in vivo*. In this study

we explore how the short-delay movement feedback from intraspinal proprioceptors to V2a interneurons plays a role in 1) timing rhythm-generation for locomotion in adult zebrafish; 2) entraining the locomotor network to rhythmic environmental cues; and 3) adapting the motor output to physical perturbations. We first show that V2a interneurons receive rhythmic, sensory-mediated midcycle inhibition in the moving animal that contributes to early burst termination and allows for higher swim frequencies compared to swimming in the absence of movement. Next, we applied controlled rhythmic bending of the body during fictive swimming and demonstrate that this entrains ongoing motor output to match the frequency of body bending. Using patch-clamp recordings of V2a interneurons we show that this entrainment is mediated by a rhythmic inhibitory signal that is sufficient to time the activity of premotor excitation to consequently entrain the whole locomotor network. We also applied an acute mechanical perturbation of the body during fictive swimming by briefly bending the body to one side. This resulted in an immediate asymmetry in motor output to counteract the perturbation, reflected by an increase in the duty cycle of motor bursts on the side opposite the bend, and decrease in duty cycle on the same side as the bend. We also show that V2a interneurons on the weakened side receive a tonic sensory inhibition that reduces their firing, which in turn leads to diminished excitation to motoneurons and hence weaker motor output. Conversely, both V2a interneurons and MNs on the augmented side are disinhibited, leading to more spikes and stronger output. Finally, we recapitulate these findings using a computational model of the sensorimotor circuit for locomotion in adult zebrafish. This model will allow us to test new hypotheses that are more difficult to test experimentally, such as the role of intraspinal sensory feedback for motor dynamics in groups of fish and the motor consequences of hydrodynamic perturbations.

Disclosures: L. Picton: None. **D. Madrid-Pulgarin:** None. **A. Pazzaglia:** None. **J. Patrick Arreguit O'Neill:** None. **A. Ferrario:** None. **A. Ijspeert:** None. **A. El Manira:** None.

Poster

PSTR157. Connectivity

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR157.06/GG5

Topic: E.09. Motor Neurons and Muscle

Support: NINDS, NIH (R01) grant NS078375
NINDS, NIH (R01) grant AA027079-05
The SMA Foundation
Project ALS

Title: Classical complement-dependent and independent mechanisms in the elimination of inappropriate synapses of sensory-motor circuits.

Authors: *D. M. FLOREZ-PAZ, G. Z. MENTIS;
Ctr. for Motor Neuron Biol. and Disease, Depts. of Neurol. and Pathology & Cell Biol., Columbia Univ., New York, NY

Abstract: Normal movement requires the proper assembly and function of sensory-motor circuits in the spinal cord. Although their assembly transpires during embryonic development, their development and refinement occur postnatally. Proprioceptive sensory neurons form synapses only with homonymous and synergistic motor neurons in mature circuits. Yet, previous reports suggested the presence of inappropriate proprioceptive synapses on motor neurons from antagonistic muscles during embryonic development. Here, we demonstrate that inappropriate synapses are initially formed and subsequently eliminated. We also shed light on the molecular mechanisms involved in this process. We previously reported that C1q, the initiating protein of the classical complement cascade, may be involved in the elimination of excessive proprioceptive synapses during postnatal development. However, is unknown whether this synaptic elimination impacts the refinement of sensory-motor circuits including the elimination of inappropriate synapses. To address this, we studied the role of C3, a downstream protein in the classical complement, known as “eat me signal” for synapse removal. Using virally-mediated map strategies together with genetic removal of C3 in mice, we found a broad increase of proprioceptive synapses and confirmed the incidence of inappropriate proprioceptive synapses on motor neurons from antagonistic muscles. Using the spinal cord-hindlimb preparation *ex vivo* and patch clamp intracellular recordings we verified that these synapses were functional. To investigate whether synapses that are destined to be eliminated can be protected, we tested whether CD47 - a protein reported as a “don’t eat me signal” in brain circuits - can confer any synaptic protection. Strikingly, genetic removal of CD47 in mice, had a similar effect to that observed in $C3^{-/-}$ mice, indicating that CD47 plays a different and unexpected role to that reported in the developing brain, acting as an “eat me signal” for synapses destined to be removed. Taken together, our results demonstrate that inappropriate synapses are eliminated in sensory-motor circuits through classical complement-dependent and independent molecular mechanisms during early spinal cord development.

Disclosures: D.M. Florez-Paz: None. **G.Z. Mentis:** None.

Poster

PSTR157. Connectivity

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR157.07/GG6

Topic: E.09. Motor Neurons and Muscle

Support: NHMRC (National Health & Medical Research Council Australia) APP1188169
University of Queensland Research Training Program Tuition Fee Offset
University of Queensland Research Training Program Stipend
Daniel McLoone MND Research Grant (IG 2326)

Title: Microglia and astrocyte mediate breakdown of perineuronal nets surrounding motor neurons of $SOD1^{G93A}$ ALS model mice

Authors: *S. CHEUNG[1,2], P. G. NOAKES[2], M. C. BELLINGHAM[2], D. G. SIMMONS[2];
[2]Sch. of Biomed. Sci., [1]Univ. of Queensland, St Lucia, Australia

Abstract: Perineuronal nets (PNNs) are extracellular matrix structures that encase excitable neurons such as motor neurons and their proximal dendrites. These nets play an important role in neuroprotection against oxidative stress. PNNs have also shown to be decreased in the ventral horn of the spinal cord of amyotrophic lateral sclerosis (ALS) rodent models. ALS is a neurodegenerative disease characterized by motor deficit symptoms and progressive muscle weakness. The loss of lower motor neurons in the ventral horn of the spinal cord is a pathological hallmark of ALS. To date, no study has defined the timeline of PNN breakdown around lower motor neurons in an ALS mouse model. We therefore investigated PNN breakdown and the possible contributing factors in the transgenic $SOD1^{G93A}$ mouse (B6-Cg-Tg (SOD1-G93A) 1Gur/J) expressing high copy numbers of mutant human SOD1 on a C57BL/6J background. These mice and their age-matched wildtype controls were investigated at four stages defined in ALS, pre-symptomatic (postnatal day 30 [P30]), onset (P70), mid-stage (P120) and end-stage (P150). Sex differences were not assessed, however, a similar ratio of male and females were used for both groups. To account for variability, six to eight mice and three technical replicates were used at each stage per genotype. Lumbar spinal cord sections were stained with antibodies and imaged using a widefield or laser scanning confocal microscope. All staining and imaging procedures were standardized between the two genotypes. A negative control for the primary antibody was used in each experiment. The experimenter was also blinded to the animal genotype. Significant breakdown of PNNs, as analysed by changes in normalized fluorescence intensity, around lower motor neurons was observed at the onset and mid-stage disease in $SOD1^{G93A}$ mice compared to wildtype controls. This was accompanied with an increase in the expression of matrix metallopeptidase-9 (MMP-9), an endopeptidase involved in degrading PNNs, in microglia and astrocytes. Microglia and astrocytes were recruited towards motor neurons in the ventral horn, and were seen to engulf PNN components. Following PNN breakdown, lower motor neurons showed increased expression of 3-nitrotyrosine, a marker for protein oxidation. Our working model suggests that recruited glia and their secreted MMP-9 mediates the breakdown of PNNs around lower motor neurons in the $SOD1^{G93A}$ mouse. The loss of these neuroprotective nets may lead to oxidative damage accumulation and lower motor neuron neurodegeneration.

Disclosures: S. Cheung: None. **P.G. Noakes:** None. **M.C. Bellingham:** None. **D.G. Simmons:** None.

Poster

PSTR157. Connectivity

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR157.08/GG7

Topic: E.09. Motor Neurons and Muscle

Title: Serotonin can beneficially compensate for the loss of the C-boutons in a mouse model of amyotrophic lateral sclerosis

Authors: *T. L. WELLS, R. POPOLI, T. AKAY;
Med. Neurosci., Dalhousie Univ., Halifax, NS, Canada

Abstract: Amyotrophic Lateral Sclerosis (ALS) is an adult-onset neurodegenerative disease with progressive motor neuron (MN) death, where patients usually die within five years of diagnosis. Since surviving MNs can compensate for those that have died during the disease, symptoms appear only after significant MN death. The C-boutons are cholinergic synapses on MNs that regulate MN excitability. Genetic inactivation of the C-boutons in $mSOD1^{G93A}$ ALS model mice ($mSOD1^{G93A}$; Dbx1::cre; $ChAT^{fl/fl}$) induces an earlier onset of symptoms. In pair with frequent swimming, however, genetic inactivation of the C-boutons has an opposite effect. Because of its role in regulating MN excitability, we hypothesized that the serotonergic system is recruited by frequent swimming to offset the inactivated C-boutons. Here, we sought be better understand this relationship. By histologically assessing c-Fos expression as a marker for neuronal activity in $mSOD1^{G93A}$ ALS model mice, we show that C-bouton activity increases at symptom onset and remains increased during symptomatic stages of the disease (ANOVA $p<0.05$). The $V0_c$ interneurons (the source of the C-boutons) also die as the disease progresses, which parallels MN death (ANOVA $p<0.01$). Serotonergic neurons increase in activity only during symptomatic stages (ANOVA $p<0.001$), indicating that they compensate for the disease later than the C-boutons. Genetic inactivation of the C-boutons induces activation of these serotonergic neurons at symptom onset when the C-boutons would normally be active (ANOVA $p<0.001$). These results indicate that different neural populations are likely recruited in a progressive manner to compensate for MN death. In pair with our previous findings, the serotonergic system can likely compensate for the loss the C-boutons in a beneficial manner. The C-boutons, serotonergic system, and other systems involved in this compensation may be potent therapeutic targets in the disease.

Disclosures: T.L. Wells: None. **R. Popoli:** None. **T. Akay:** None.

Poster

PSTR157. Connectivity

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR157.09/GG8

Topic: E.07. Rhythmic Motor Pattern Generation

Support: NIH Grant R01NS112304
NIH Grant R01NS089324
NIH Grant R01NS115900

Title: Reorganization of spinal neural connectivity following recovery after thoracic spinal cord injury: insights from computational modeling

Authors: N. A. SHEVTSOVA[1], I. A. RYBAK[1], D. S. MAGNUSON[2], ***S. M. DANNER**[1]; [1]Neurobio. and Anat., Drexel Univ., Philadelphia, PA; [2]Dept. of Neurolog. Surgery, Univ. of Louisville, Louisville, KY

Abstract: During overground locomotion, intact rats express alternating (walk and trot) and non-alternating (canter, gallop, half-bound gallop, and bound) gaits in a speed-dependent manner. After spinal cord injury (SCI), such as thoracic lateral hemisection or moderate contusion, rats recover the ability to locomote, however with altered gait expression and reduced maximal and average speed and stepping frequency. Specifically, after recovery from hemisection, rats express only a subset of intact gaits (they do not use the highest-speed gaits half-bound gallop and bound) and predominantly use the limb contralateral to injury side as lead during canter and gallop. A mild-moderate midline contusion injury causes a greater reduction in maximal speed, loss of all non-alternating gaits, and emergence of novel alternating gaits. These changes likely result from the reorganization of the spared and injured spinal circuitry, including long propriospinal neuronal (LPN) connections between the cervical and lumbar spinal cord enlargements crucial for forelimb and hindlimb coordination. To investigate possible reorganization of neural connectivity between and within the cervical and lumbar enlargements following post-SCI recovery, we built upon our previous model of spinal circuitry that included the cervical and lumbar compartments with rhythm generating circuits for each limb (RGs) interacting through commissural interneurons (CINs) within each compartment and both ascending and descending LPNs providing communication between the compartments. Locomotor-like behavior in our model was produced by simulated brainstem drives to RGs, CINs, and LPNs. The contusion and hemisection were simulated by varying brainstem drive to and connectivity between and within the two compartments. Our model was able to reproduce the experimentally observed speed-dependent gait expression for both intact and injured rats, including altered post-injury gait expression. The model suggests that following contusion, strengthened intra-enlargement connectivity and descending drives compensated for the weakened LPN connections. For hemisection, the model required substantial recovery of the severed LPN and descending drives to function suggesting either strengthening of homo- and heterolateral connections or formation of detour pathways via the intact hemicord to compensate for the unilateral injury.

Disclosures: N.A. Shevtsova: None. **I.A. Rybak:** None. **D.S. Magnuson:** None. **S.M. Danner:** None.

Poster

PSTR157. Connectivity

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR157.10/GG9

Topic: E.07.a. Cellular properties – Interneurons and motor neurons

Support: NSF Grant IOS1754869

Title: Intracellular Neural Control of Aplysia Feeding in a Semi-intact Preparation

Authors: *Y. HUAN[1], B. N. TIBBETTS[1], P. R. PATEL[2], C. A. CHESTEK[2], H. J. CHIEL[1];
[1]Case Western Reserve Univ., Cleveland, OH; [2]Univ. of Michigan, Ann Arbor, MI

Abstract: Adaptive behavior is important for animals' survival. To adapt to changing environments, flexibility within a neural circuit is required so that the same neural network can produce different motor responses. In *Aplysia*, feeding behaviors are produced by a neural circuit located in the buccal and cerebral ganglia. This feeding circuit can generate different rhythmic patterns to ingest or egest food by recruiting different neurons and regulating the activity of those neurons (Lu et al., 2013). To study the neural circuit mechanism of multifunctionality, the ability to both precisely control the neural circuit while monitoring behavior is beneficial. We used a carbon fiber array on a semi-intact preparation to monitor and manipulate *Aplysia*'s feeding circuit. During the experiments, the cerebral ganglia, the buccal ganglia, and the feeding apparatus (buccal mass) were isolated. A 16-channel platinum-iridium coated carbon fiber array (each fiber is ~8 µm in diameter) was inserted into a desheathed buccal ganglion to record and stimulate neurons intracellularly. Carbachol solution (~5 mM) was applied to the cerebral ganglia to elicit feeding behaviors in the buccal mass. We were able to record intracellularly from multiple neurons as the buccal mass vigorously ingested food. By injecting current through the carbon fiber electrodes, we stimulated an individual motor neuron (B3) to elicit contraction of the musculature controlling the jaws (I3 muscle) and inhibited an individual multi-action neuron (B4/B5) to interrupt a feeding movement. Our preliminary data showed that we could use carbon fiber arrays to control individual neurons in a behaving buccal mass. This method allows us to monitor the intracellular activity from multiple neurons simultaneously during real feeding movements, providing information about the neural connectivity during different feeding behaviors. The ability to control the neurons also provides a new opportunity to study how a neural circuit generates different feeding behaviors and how adaptive behaviors can be modified by manipulating inputs into a neural circuit.

Disclosures: Y. Huan: None. **B.N. Tibbetts:** None. **P.R. Patel:** None. **C.A. Chestek:** None. **H.J. Chiel:** None.

Poster

PSTR157. Connectivity

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR157.11/GG10

Topic: E.07.a. Cellular properties – Interneurons and motor neurons

Support: NINDS Grant NS047073
NSF Grant DMS-1010434
NSF Grant IIS-1065489

Title: Regionally specific activation mediates a wave of contraction within Aplysia's retractor muscle through actions of identified neurons

Authors: *J. M. MCMANUS, H. LU, H. J. CHIEL;
Biol., Case Western Reserve Univ., Cleveland, OH

Abstract: The muscles of animals are often not single functional units, but instead can be activated differentially across different regions. How is this regional activation controlled by nervous systems, and what is its behavioral significance? Regional differences within muscles have been described in many vertebrate systems, but fewer studies have explored this phenomenon at the identified neuron level. The I3 muscle of the marine mollusk *Aplysia* is innervated by a pool of identified motor neurons with different innervation regions, so regional differences in muscle activation can be tied to identified neurons. A previous study showed that the anterior region of I3, the primary retractor of *Aplysia*'s food grasper, has an additional function during the protraction phase of swallowing behaviors: holding food. This function was shown to occur through activity of identified motor neuron B38. A second motor neuron, B39, is also known to specifically activate I3's anterior. What is the behavioral function of B39? To address this question, we measured forces induced in different regions of I3 both during *in vitro* motor programs and in response to intracellular stimulation of B39, and we recorded B39's activity during different motor programs. We also used a semi-intact preparation capable of feeding movements to observe muscle contractions and corresponding neural activity in different motor programs. We found that, at the onset of the retraction phase in rejections but not in other behaviors, I3's anterior contracts prior to contraction of the whole muscle, resulting in an anterior to posterior wave of contraction. We additionally found that B39 is specifically activated in these patterns. Our results also suggest an interneuronal basis for the wave of contraction within I3: identified neurons B4/B5, which are intensely active in rejection, inhibit motor neurons that activate the other I3 muscle regions but do not inhibit B39. During rejections, *Aplysia*'s food grasper retracts while open to release inedible material. An anterior to posterior wave of contraction in the retractor muscle I3 could help push the open grasper inward without squeezing it closed on the inedible material, which was previously shown could occur with premature activation of the whole I3 during rejection. Additionally, the anterior I3 via B39 in rejection could have a similar grasping function as it does via B38 in swallowing, at a different phase of the motor program. Our results help illuminate, at the level of individual, identified neurons, how an animal takes advantage of regionally specific muscle activation to generate multiple qualitatively different behaviors using the same musculature.

Disclosures: J.M. McManus: None. **H. Lu:** None. **H.J. Chiel:** None.

Poster

PSTR157. Connectivity

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR157.12/GG11

Topic: E.07.a. Cellular properties – Interneurons and motor neurons

Support: NIH-NINDS 1R15NS128619-01
NSF-IOS 1755283

Title: Dual-network regulation of motor neuron activity modifies muscle electrical response

Authors: *B. GNANABHARATHI, L. A. ZEIGLER, A. R. PINTER, A. A. SHERIDAN, S.-R. H. FAHOUM, D. M. BLITZ;
Miami Univ., Oxford, OH

Abstract: Neuronal participation in multiple networks can mediate coordination of related behaviors (e.g. vocalizing, breathing, locomoting) (Schmidt et al, Exp Physiol. 2012). However, the extent of dual-network activity, and thus coordination, differs across modulatory states (Juvin et al, Front Neuroanat 2022), so it is important to understand how multi-network activity influences muscles generating the coordinated behavior. Here we explore how dual-network participation, and inter-network interactions, influence muscles seemingly dedicated to one behavior. We record intracellularly from single muscle fibers in nerve-muscle preparations from the crab (*Cancer borealis*) stomatogastric system, which includes two well-defined rhythmic networks (gastric mill: ~0.1 Hz, chewing; pyloric: ~1 Hz, filtering food). The "SIF" modulatory state (modulatory neuron MCN5/Gly^1-SIFamide neuropeptide) switches the pyloric-only LPG neuron into dual pyloric/gastric mill network participation (Fahoum & Blitz, J Neurosci 2021). LPG innervates both pyloric (p7) and gastric mill (gm3b,c) muscles. Synaptic and cellular properties can have complex effects on muscle responses to neural input (Morris et al Nat Neurosc 2000; Blitz et al, J Exp Biol. 2017; Daur et al eNeuro 2021). We hypothesize that LPG dual-network activity differently impacts its pyloric and gastric mill muscle targets. We used high, medium and low intensity stimulation patterns based on recorded LPG dual activity and recorded p7 excitatory junction potentials (EJPs). p7 EJPs followed dual-network activity for each pattern with larger amplitudes maintained during gastric mill bursts in medium/high vs. low patterns ($N = 7$ animals; $n = 8$-10 muscle fibers, $p<0.01$). Also in this SIF modulatory state, the cycle period of LPG pyloric activity is phasically prolonged by gastric mill network input (Gnanabharathi et al, SfN abstract 2022). Comparisons to stimulation patterns with prolongations eliminated revealed an increased EJP amplitude persisted for two pyloric cycles after a prolongation ($N = 1$; $n = 7$; $p<0.02$ - 0.004). Further, the EJP_2/EJP_1 ratio was increased for the next two pyloric bursts after a prolongation ($p<0.001$-0.04). Therefore, p7 activity appears sensitive to LPG dual-network activity and inter-network effects. Future goals are to test if LPG gastric mill muscle gm3b,c properties prohibit dual-network activity, and if the influence of dual-network activity at the muscle level varies across modulatory states. Overall, the complexity of the neuromuscular transform appears to shape the influence of interacting networks, thus participating actively in the coordination of related behaviors.

Disclosures: B. Gnanabharathi: None. **L.A. Zeigler:** None. **A.R. Pinter:** None. **A.A. Sheridan:** None. **S.H. Fahoum:** None. **D.M. Blitz:** None.

Poster

PSTR158. Molecular and Cellular Changes with Reproductive State and Hormones

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR158.01/GG12

Topic: C.01. Brain Wellness and Aging

Support: •Jacqueline Ford Gender and Health Fund (to G.E.)
•Canadian Consortium on Neurodegeneration in Aging (CCNA) Phase II (grant CCNA 049-04 to H.Chertkow, G.Einstein et al.) i.CIHR reference number: CNA 163902
Wilfred and Joyce Posluns Chair in Women's Brain Health and Aging from the Posluns Family Foundation, Women's Brain Health Initiative, Canadian Institutes of Health Research (CIHR reference no. WJD-180960) to GE.

Title: Cognitive health in middle-aged women with different type of menopauses: implications for risk, resilience and subjective cognitive decline

Authors: *N. CALVO[1], L. GRAVELSINS[1], A. BROWN[1], S. ZHAO[1], S. RAMANA[1], G. EINSTEIN[1,2,3,4,5];
[1]Univ. of Toronto, Toronto, ON, Canada; [2]Rotman Res. Institute, Baycrest Hlth. Sci., Toronto, ON, Canada; [3]Tema Genus, Linköping Univ., Linköping, Sweden; [4]Women's Col. Res. Inst., Toronto, ON, Canada; [5]Ctr. for Life Course and Aging, Univ. of Toronto, Toronto, ON, Canada

Abstract: Understanding risk and resilience proxies in middle-aged women is crucial because they might influence the progression from normal aging to Subjective Cognitive Decline (SCD); and thus, provide evidence for early detection of dementia. Midlife and menopause represent major physiological shifts in a woman which are typically accompanied by molecular and neuroplastic adaptations. For instance, during spontaneous menopause (SM) women have declining 17-b-estradiol (E2) levels; a hormone that prevents neuronal death, shapes hippocampal plasticity, and affects cognitive and brain reserve. While SM, itself, may be a risk factor, there are different types of menopauses including oophorectomy which occurs on the average of 10 years younger than SM and is accompanied by an immediate loss of ovarian E2 synthesis. These different menopauses may lead to cognitive changes, and differentially affect brain regions subserving memory. We explored risk and resilience factors in a cohort of 360 middle-aged women with neuropsychological testing and MRI. The cohort included younger-middle-aged women with the BReast CAncer mutation (BRCA1/2) with and without prophylactic bilateral salpingo-oophorectomy (BSO), age-matched premenopausal women without BRCA1/2 and with ovaries (AMC), and women in SM- any of whom might be *APOE ε4* carriers. As a first step, we conducted a Partial Least Squares Correlation Analysis (PLSC) in the full sample which recognized one latent variable (LV) that explained 73% of the co-variance. This LV indicated that BRCA 1, poor mood, higher age, *APOE ε4*, higher stress, and BSO are associated with decreased performance on neuropsychological measures (e.g., memory, language, attention). Thus, these variables may be risk factors for cognitive decline in this cohort. Alternatively, higher education, verbal IQ, taking hormonal contraceptives, and multilingualism were associated with increased performance, indicating that these might contribute to resilience. Across the groups, some of these middle-aged women reported SCD (n=113), and the main risk factors in these women were BSO, BRCA1 and *APOE ε4*; while the main resilience factors were education, verbal IQ and higher E2 level. Moreover, factorial analyses revealed that low E2 (either BSO or spontaneous menopause) may lead to decline in

verbal episodic and spatial working memory together with decreases in brain structure. These results suggest that risk and resilience factors affect middle-aged women differently depending on type of menopause, E2 levels, genotype and SCD status.

Disclosures: **N. Calvo:** None. **L. Gravelsins:** None. **A. Brown:** None. **S. Zhao:** None. **S. Ramana:** None. **G. Einstein:** None.

Poster

PSTR158. Molecular and Cellular Changes with Reproductive State and Hormones

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR158.02/GG13

Topic: F.02. Neuroendrocrine Processes and Behavior

Support: Discovery Grant from NSERC

Title: The Effects of Hormonal Contraceptives on Dendritic Spine Density in the CA1 Region of the Hippocampus

Authors: *E. GOMEZ-PERALES[1], I. FERRAZ[2], E. BUCHANAN[1], L. M. BUYNACK[1], L. E. ERNEDAL[1], E. F. GANNÉ[1], L. C. DE MELO RODRIGUES[2], W. G. BRAKE[1];
[1]Concordia Univ., Montreal, QC, Canada; [2]Univ. Federal do Espírito Santo, Vitória, Brazil

Abstract: Dendritic spine density in the CA1 region of the hippocampus fluctuates throughout a rat's estrus cycle. This change in dendritic spine density has been associated with circulating levels of estrogens and progesterone. Administration of ethinyl estradiol (EE) and levonorgestrel (LNG), which are synthetic hormones found in a commonly used formulation of hormonal contraceptives, disrupts the estrus cycle. Here we investigate how these synthetic hormones affect dendritic spine density in the CA1 region of the hippocampus and contrast these effects with rats in three phases of the estrus cycle. Gonadally-intact Long-Evans female rats were assigned to 6 groups (N=36). There were three cycling groups; Proestrus, Estrus, Diestrus - and three hormone treatment groups; EE (10 μg/kg), LNG (20 μg/kg) EE+LNG (10+20 μg/kg). They received daily subcutaneous injections of either sesame oil (for the cycling groups), or EE, LNG, or EE+LNG for 21 days and the phase of their estrus cycle was tracked daily using vaginal cytology. The brains were collected and stained according to the FD Rapid Golgi Stain protocol. Hippocampal slices were cut at 100 μm thickness and analyzed using light microscopy at 60x magnification. Dendritic spines were counted and classified using Neurolucida software. Preliminary data suggests that female rats in proestrus exhibited a greater mature dendritic spine density compared to rats in estrus. Female rats in the EE+LNG group had a greater mature dendritic spine density compared to rats receiving EE or LNG alone. In contrast, when looking at the total number of spines, rats receiving EE alone exhibited lower total dendritic spine density compared to rats receiving LNG alone or the EE+LNG group. Phases of the estrus cycle did not affect total dendritic spine density in naturally cycling female rats. These preliminary data

suggest that hormonal contraceptives affect dendritic spine density in female rats and their effect depends upon the formulation of the hormonal contraceptive.

Disclosures: E. Gomez-Perales: None. **I. Ferraz:** None. **E. Buchanan:** None. **L.M. Buynack:** None. **L.E. Ernedal:** None. **E.F. Ganné:** None. **L.C. de Melo Rodrigues:** None. **W.G. Brake:** None.

Poster

PSTR158. Molecular and Cellular Changes with Reproductive State and Hormones

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR158.03/GG14

Topic: F.02. Neuroendrocrine Processes and Behavior

Support: R37HD034860
1F31HD110102-01

Title: Roles for hypothalamic astrocytes in neural circuits controlling reproduction

Authors: *C. D. PHILLIPS, R. A. DEFAZIO, S. M. MOENTER;
Mol. & Integrative Physiol., Univ. of Michigan, Ann Arbor, Ann Arbor, MI

Abstract: Astrocytes are critical for many central nervous system functions. Disrupting hypothalamic astrocytes can impair reproduction. Gonadotropin-releasing hormone (GnRH) neurons in the preoptic area (POA) and hypothalamus produce the final central output controlling fertility. An afferent network likely governs GnRH secretion, including both astrocytes and kisspeptin neurons in the arcuate nucleus (KNDy neurons; ARC). Activating Gq signaling in POA astrocytes using chemogenetics initiates GnRH neuron firing in brain slices from male mice, but KNDy neurons were unresponsive. We tested if factors including sex, female reproductive cycle stage, brain region, and time of day affect astrocyte regulation of reproduction. We targeted glial fibrillary acidic protein-driven expression of the hM3Dq designer receptor exclusively activated by designer drug (DREADD) to astrocytes in specific regions by stereotaxic injection of AAVs. We used Ca^{2+} imaging to verify that application of the DREADD ligand clozapine N-oxide (CNO) induces Ca^{2+} signaling in mouse brain slices. We found that CNO induces Ca^{2+} signaling in both POA and ARC astrocytes regardless of sex, the time of day of recording (morning [AM] v. afternoon [PM]), or cycle stages (diestrus v. proestrus). To examine if astrocyte stimulation changes neuroendocrine parameters, we carried out *in vivo* serial blood sample collections assayed for luteinizing hormone (LH), which serves as a bioassay for GnRH release, as well as extracellular recordings of GnRH and KNDy neurons in brain slices. Activating POA astrocytes with CNO increased LH levels in both males and diestrous females regardless of the time of day of sampling, but the rate of LH increase in males was faster than in females in both the AM and PM (both times $p < 0.001$ at 10 min, 3-way ANOVA). Further, LH release in males was more sustained than in females in the AM and PM (both times $p < 0.001$, 3-way ANOVA). Activating ARC astrocytes with CNO induced a small increase in

LH levels in males, but not females. Extracellular recordings showed GnRH neuron firing rates increasing following POA astrocyte stimulation with CNO regardless of sex, the time of day of recording, and cycle stage, but ARC astrocyte stimulation had no effect on KNDy neuron firing rates in any group. Our findings provide evidence of POA astrocyte regulation of GnRH neurons and subsequent LH release in both sexes regardless of time of day; mechanisms of the delayed response in females need further study. In contrast, ARC astrocytes in males, but not females, signal via an unknown population to increase LH. An intriguing possibility is a sex difference in the response of GnRH neuron projections to gliotransmitters.

Disclosures: C.D. Phillips: None. **R.A. DeFazio:** None. **S.M. Moenter:** None.

Poster

PSTR158. Molecular and Cellular Changes with Reproductive State and Hormones

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR158.04/GG15

Topic: F.02. Neuroendrocrine Processes and Behavior

Support: Psychopharmacology Research Fund

Title: Activity, nociception, and morphine antinociception in a female-to-male transgender model in the rat

Authors: *R. CRAFT[1], **C. SEWELL**[2], T. TAYLOR[2], M. VO[2], M. M. MORGAN[2]; [1]Washington State Univ., Pullman, WA; [2]Washington State Univ., Vancouver, WA

Abstract: Gender-affirming testosterone therapy has been reported to decrease the experience of pain in some transgender men. Our objective was to develop a female to male transgender model in rats to test this hypothesis. Adult female Sprague-Dawley rats were implanted subcutaneously with a continuous release testosterone capsule followed 3 weeks later by removal of the ovaries. Control rats received a blank placebo capsule and sham surgery. Treating females with testosterone eliminated estrous cycling, increased body weight, and decreased uterine weight. Testosterone also decreased home cage wheel-running compared to controls. Subsequent ovariectomy, which decreased wheel running in placebo-treated females, did not alter activity in testosterone-treated females. Injection of CFA into the hindpaw to induce inflammatory pain decreased wheel running in all rats. Neither pain-depressed running nor the antinociceptive effect of morphine was significantly altered by testosterone or ovariectomy, although there was a trend for testosterone to enhance pain-depressed running and for ovariectomy to reduce the antinociceptive effect of 1 mg/kg morphine. Administration of 3.2 mg/kg morphine produced antinociception in all groups on the von Frey test while simultaneously depressing wheel running. Changes in estrous cycle, uterine and body weight, and wheel running caused by continuous testosterone administration are consistent with transition to a male phenotype. Contrary to retrospective reports of reduced pain in transgender men, no pronounced changes in

nociception or morphine antinociception were evident in female rats treated with testosterone, with or without ovariectomy.

Disclosures: **R. Craft:** None. **C. Sewell:** None. **T. Taylor:** None. **M. Vo:** None. **M.M. Morgan:** None.

Poster

PSTR158. Molecular and Cellular Changes with Reproductive State and Hormones

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR158.05/GG16

Topic: F.02. Neuroendrocrine Processes and Behavior

Support: R21DK127296
K01HL153205-01

Title: Loss of microglia inflammatory signaling in female mice exacerbates the impairment of reproductive function induced by high fat diet feeding.

Authors: *I. VELASCO[1], T. Z. JAFARI[2], J. MUMMA[2], A. L. EVANS[2], R. YOAKUM[2], J. M. FREY[1], O. SANTIAGO[1], J. P. THALER[3], M. D. DORFMAN[1];
[1]UW Med. Diabetes Institute, Univ. of Washington, Seattle, WA; [2]Univ. of Washington, Seattle, WA; [3]Metabolism, Endocrinol. & Nutr., Univ. of Washington, Sch. of Med., Seattle, WA

Abstract: Reproductive function and energy metabolism are tightly connected, but the mechanisms linking them are not completely elucidated. Microglia, the resident immune cells of the brain, are key mediators of susceptibility to diet-induced obesity (DIO). Rodents exposed to high-fat diet (HFD) develop microglial inflammation (gliosis) in the mediobasal hypothalamus (MBH) prior to significant weight gain, and microglial-specific deletion of the inflammatory NF-κB pathway prevents gliosis and DIO. Microglia activation has also been implicated in polycystic ovarian syndrome (PCOS) pathogenesis and the regulation of GnRH neurons, suggesting a role for microglial inflammatory signaling in reproductive function. To explore this possibility, we studied female mice with microglia-specific ablation of IKKβ (IKKβ-MGKO), a critical regulator of the NF-κB pathway. Adult wild-type (WT) and IKKβ-MGKO female mice were fed regular chow or a HFD for 12 weeks. Compared with chow-fed mice, IKKβ-MGKO females fed a HFD had increased hypothalamic expression of essential genes involved in control of the hypothalamus-pituitary-gonadal (HPG) axis such as NKB, Kiss1 and its receptor Kiss1r. Accordingly, IHC analysis revealed a significant increase in NKB protein levels in the MBH of HFD-fed IKKβ-MGKO mice. Interestingly, HFD feeding disrupted estrous cyclicity in 47% of IKKβ-MGKO mice compared with 25% of WT females, suggesting a protective function of microglial inflammatory signaling. Furthermore, KO females on HFD displayed enlarged ovaries with reduced numbers of corpora luteum, accompanied by lower expression of aromatase, LH receptor and Kiss1r in the ovary. Together, these KO phenotypes resemble pathologies in which hyperstimulation of the reproductive axis causes ovarian dysfunction (e.g. PCOS), supporting the

hypothesis that microglial inflammatory signaling helps preserve female reproductive function during DIO.

Disclosures: I. Velasco: None. **T.Z. Jafari:** None. **J. Mumma:** None. **A.L. Evans:** None. **R. Yoakum:** None. **J.M. Frey:** None. **O. Santiago:** None. **J.P. Thaler:** None. **M.D. Dorfman:** None.

Poster

PSTR158. Molecular and Cellular Changes with Reproductive State and Hormones

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR158.06/GG17

Topic: F.02. Neuroendrocrine Processes and Behavior

Support: NIGMS Grant RL5GM118972

Title: A computational model for the excitability of hypothalamic neuronal circuitry during pregnancy

Authors: M. BANDORA[1], P. BENNETT[2], J. COOPER[3], C. PATIN[4], C. F. ELIAS[5], ***P. LEE**[3]; [1]Biol. and Computer Sci., [2]Computer Sci., [3]Mathematics, Morgan State Univ., Baltimore, MD; [4]Mol. and Integrative Physiol., Univ. of Michigan, Ann Arbor, MI; [5]Mol. and Integrative Physiol., Univ. of Michigan Neurosci. Grad. Program, Ann Arbor, MI

Abstract: Energy sensing in reproduction is mainly modulated by the ventral premammillary nucleus (PMv) neurons in hypothalamus. During pregnancy, elevated prolactin hormone induces leptin resistance and increase in body weight. At the same time, dopamine released from tuberoinfundibular dopaminergic (TIDA) neurons is downregulated. PMv neurons express prolactin receptors and might interplay with leptin. Even though leptin action in PMv neurons has not been associated with metabolic regulation, very little is known about the potential role in physiologic states of negative energy balance during pregnancy.
The neuronal population sensitive to the adipocye-derived hormone leptin (LepRb PMv) is not homogeneous. A subset of LepRb PMv neurons depolarizes, whereas another hyperpolarizes in response to leptin. We have identified for adult females, that 1) PMv DAT neurons project densely to Kisspeptin neurons in the anteroventral periventricular nucleus (AVPV), 2) minimal projection to Kisspeptin neurons in the arcuate nucleus (ARC) or AgRP neurons, (3) DAT, LepRb, and prolactin receptor (PrlR) are coexpressed in PMv neurons.
We hypothesize that 1) the populated PMv DAT neurons sensing upregulated prolactin innervate AgRP axonal terminals for stronger inhibition of Kiss1 neurons in the ARC in the stage of pregnancy, 2) the difference in DAT mRNA expression between adult and pregnant mice is a direct signature that PMv-LepR neurons display gestational plasticity associated with the metabolic control of pregnancy.
Accordingly, we have developed a computational model of conductance-based electrophysiology to support these hypotheses. Input signals to PMv and AgRP (agouti-related peptide) neurons by

leptin, and the output response of bursting spikes and their relays to Kiss1 neurons in the AVPV and the ARC, and ultimately to GnRH neurons is assessed. Among PMv neurons, the primary ion channels are identified by single-cell RNA-seq data considering subpopulations of LepR neurons, i.e., DAT+ and DAT-.
The computational neuronal circuit model predicts that the contribution of upregulated prolactin and DAT are significant for the suppression of GnRH pulsatility, as a signature of an adaptation in the negative energy balance.

Disclosures: **M. Bandora:** None. **P. Bennett:** None. **J. Cooper:** None. **C. Patin:** None. **C.F. Elias:** None. **P. Lee:** None.

Poster

PSTR158. Molecular and Cellular Changes with Reproductive State and Hormones

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR158.07/GG18

Topic: F.02. Neuroendrocrine Processes and Behavior

Support: Nagoya University Interdisciplinary Frontier Fellowship Grant JPMJFS2120
KAKENHI Grants 19H03103
KAKENHI Grants 20H03127
KAKENHI Grants 18H03973, 21H05031

Title: Purinergic signaling directly activates AVPV kisspeptin neurons via P2X2 receptor to induce GnRH/LH surge and ovulation in female rats

Authors: *S. HAZIM[1], S. SEKI[1], M. NAGAE[1], T. HITOMI[1], R. YABUSHITA[1], M. HIRABAYASHI[2], Y. UENOYAMA[1], H. TSUKAMURA[1], N. INOUE[1];
[1]Dept. of Animal Sci., Grad. Sch. of Bioagricultural Sciences, Nagoya University, Japan, Aichi, Nagoya, Japan; [2]Ctr. for Genet. Analysis of Behavior, Natl. Inst. for Physiological Sci., Nagoya, Japan

Abstract: Ovulation disorders are a major cause of low pregnancy and infertility in humans as well as livestock. Hypothalamic kisspeptin neurons are considered key regulators of the Hypothalamus-pituitary-gonadal (HPG) axis in mammals including rodents. Kisspeptin neurons located in the anteroventral periventricular nucleus (AVPV) in rodents and preoptic area in other species are considered to be responsible for the generation of gonadotropin-releasing hormone (GnRH)/luteinizing hormone (LH) surge and consequent ovulation in female mammals. Our recent study suggested that hindbrain adenosine triphosphate (ATP)-purinergic signaling triggers LH surge and ovulation via activation of AVPV kisspeptin neurons in rats. However, it remained unclear if ATP directly activates the AVPV kisspeptin neurons via *P2rx2*, a purinergic receptor, expressed in these neurons. The present study aimed to examine the effect of the AVPV kisspeptin neuron-specific knockdown of *P2rx2* expression on the estrogen-induced LH surge in

female rats using *Kiss1-Cre* rats. AAV containing *P2rx2* shRNA or Scrambled shRNA was injected into the AVPV of *Kiss1-Cre* female rats. Two weeks later, the rats were ovariectomized (OVX) and treated with a diestrous level of estradiol (E2) for 5 days and a proestrous level of E2 (high E2) for 2 days. Then, blood samples were collected to detect high E2-induced LH surge. As a result, endogenous afternoon LH surge was significantly suppressed in *P2rx2* knocked-down OVX + high E2 rats compared to the scrambled shRNA-injected control rats. Furthermore, to determine the effects of AVPV kisspeptin neuron-specific *P2rx2* knockdown on ovulation, ovary-intact *Kiss1-Cre* female rats were injected with AAV containing *P2rx2*-shRNA or Scrambled shRNA. Three weeks after the injection, blood samples were collected to measure plasma LH levels in AAV-injected proestrus *Kiss1-Cre* rats and ovulated oocytes were counted on the next day of blood sampling. Spontaneous LH surge was significantly suppressed, and the number of ovulated oocytes decreased in AVPV kisspeptin neuron-specific *P2rx2* knocked-down proestrus rats compared to the control group. These findings suggest that the purinergic neurons directly stimulate the AVPV kisspeptin neurons via P2X2 receptor to trigger GnRH/LH surge and ovulation in female rats.

Disclosures: S. Hazim: None. **S. Seki:** None. **M. Nagae:** None. **T. Hitomi:** None. **R. Yabushita:** None. **M. Hirabayashi:** None. **Y. Uenoyama:** None. **H. Tsukamura:** None. **N. Inoue:** None.

Poster

PSTR158. Molecular and Cellular Changes with Reproductive State and Hormones

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR158.08/GG19

Topic: F.02. Neuroendrocrine Processes and Behavior

Support: JSPS KAKENHI Grant 19H03103
JSPS KAKENHI Grant 23H02362
JSPS KAKENHI Grant20H03127
JSPS KAKENHI Grant21H05031

Title: Purinergic signaling triggers LH surge and ovulation via activation of AVPV kisspeptin neurons in rats

Authors: *N. INOUE, S. HAZIM, H. TSUCHIDA, Y. OTSUKA, K. YAMADA, Y. UENOYAMA, H. TSUKAMURA;
Nagoya Univ., Nagoya, Japan

Abstract: In female rodents, kisspeptin neurons in the anteroventral periventricular nucleus (AVPV) are considered to be an estrogen action site and are responsible for generating gonadotropin-releasing hormone (GnRH)/luteinizing hormone (LH) surge and consequent ovulation. Estrogen increases *Kiss1*/kisspeptin expression in AVPV kisspeptin neurons and kisspeptin directly stimulates GnRH neurons via GPR54. Furthermore, the preovulatory levels of

estrogen activate AVPV kisspeptin neurons in rats prior to LH surge. However, upstream stimulator(s) that activate AVPV kisspeptin neurons have not been fully defined. We found that *P2rx2* is highly expressed in the AVPV kisspeptin neurons by RNA-seq analysis of visualized AVPV kisspeptin neurons taken from *Kiss1*-tdTomato heterozygous (*Kiss1*-tdTomato) female rats. The P2X2 receptor is a stimulatory cation channel gated by extracellular ATP. Thus, purinergic-P2X2-receptor signaling could be specifically involved in the activation of AVPV kisspeptin neurons. The present study aimed to investigate whether ATP-purinergic receptor signaling mediates the GnRH/LH surge generation and ovulation via activating AVPV kisspeptin neurons in female rats. Administration of a P2X receptor antagonist (PPADS) into the AVPV blocked the LH surge in ovariectomized (OVX) rats treated with a proestrous level of estradiol-17β (OVX + high E2) and significantly reduced the ovulation rate in proestrous ovary-intact rats. An administration of ATP into the AVPV induced a surge-like LH increase in OVX + high E2 rats in the morning. On the other hand, AVPV ATP administration could not induce the LH increase in OVX + high E2 *Kiss1* knockout rats. Furthermore, ATP significantly increased intracellular Ca^{2+} levels in immortalized kisspeptin neuronal cell line, and co-administration of PPADS blocked the ATP-induced Ca^{2+} increase. Histological analysis revealed that the proestrous level of E2 significantly increased the number of P2X2 receptor-immunoreactive AVPV kisspeptin neurons visualized by tdTomato in *Kiss1*-tdTomato rats. The proestrous level of E2 significantly increased varicosity-like vesicular nucleotide transporter (VNUT, a purinergic marker)-immunoreactive fibers projecting to the vicinity of AVPV kisspeptin neurons. Furthermore, we found that A1 and A2 hindbrain VNUT-positive neurons projected to the AVPV and expressed estrogen receptor α, and the neurons were activated by the high E2 treatment in female rats. These results suggest that ATP signaling from purinergic neurons in the hindbrain A1 and A2 triggers GnRH/LH surge generation and then ovulation via activation of AVPV kisspeptin neurons.

Disclosures: **N. Inoue:** None. **S. Hazim:** None. **H. Tsuchida:** None. **Y. Otsuka:** None. **K. Yamada:** None. **Y. Uenoyama:** None. **H. Tsukamura:** None.

Poster

PSTR158. Molecular and Cellular Changes with Reproductive State and Hormones

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR158.09/GG20

Topic: F.02. Neuroendrocrine Processes and Behavior

Support: NIH NIH R01HD41469
NIH F31HD097830

Title: Characterization of a central circuit controlling ovulation in mice

Authors: *J. R. STARRETT, S. M. MOENTER;
Univ. of Michigan, Ann Arbor, Ann Arbor, MI

Abstract: In spontaneously-ovulating mammals, ovulation is induced by a cascade of neuroendocrine events beginning with ovarian estradiol acting within the brain inducing a surge of gonadotropin-releasing hormone (GnRH) release from the hypothalamus. GnRH surge generation is thought to involve a population of kisspeptin-expressing neurons in the rostral hypothalamus (anteroventral-periventricular area (AVPV) in mice). Kisspeptin is a strong activator of GnRH neurons, kisspeptin expression in the AVPV positively correlates with circulating estradiol, and AVPV kisspeptin neurons exhibit increased firing activity during the GnRH surge. It has been hypothesized that priming of these cells by estradiol and activation by a diurnal signal induce GnRH surges. We used optogenetics and electrical recordings in acute brain slices to study neurotransmission between AVPV kisspeptin neurons and GnRH neurons to test the hypothesis that estradiol priming increases communication between these cells. The channelrhodopsin ChrimsonR was expressed specifically in AVPV kisspeptin neurons of female Kiss1-Cre;GnRH-GFP mice using viral (AAV) injection. Brain slices were prepared from mice in various conditions: ovariectomized (OVX, 2 or 7 days), OVX (2 days), OVX + estradiol (E) treatment, diestrous, and proestrous. Loose-patch recordings were used to measure changes in preoptic area GnRH neuron firing rate in response to photostimulation of nearby ChrimsonR-positive fibers. Stimulation (20 Hz 30s) induced delayed (30-90 s) increases in mean GnRH neuron firing rate in all groups but not all cells. After each loose-patch recording, whole-cell recording was performed to test for evoked ionotropic transmission. A similar percentage of GnRH neurons from diestrous (23%, 5 of 22), proestrous (23%, 5 of 22), and estradiol-treated ovariectomized mice (26%, 6 of 23) had evoked monosynaptic GABAergic post-synaptic currents (evPSCs). In contrast, no GnRH neurons in 2-day OVX (0 of 20) and just 8% (1 of 12) of neurons in 7-day OVX mice exhibited evPSCs, suggesting loss of ovarian hormones prunes GABAergic connectivity between AVPV kisspeptin neurons and GnRH neurons. Interestingly, despite absence of evPSCs, most (88%, 37 of 42) GnRH neurons exhibited delayed increases in firing rate, compared to 90% of neurons with evPSCs (9 of 10). This finding highlights an important consideration for “circuit-mapping” experiments. These results suggest ovarian status regulates fast-synaptic connectivity, but downstream activation of GnRH neurons remains possible across tested ovarian conditions.

Disclosures: J.R. Starrett: None. **S.M. Moenter:** None.

Poster

PSTR158. Molecular and Cellular Changes with Reproductive State and Hormones

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR158.10/HH1

Topic: F.02. Neuroendrocrine Processes and Behavior

Support: NSERC: Discovery Grant

Title: The effects of ovarian hormones on membrane progesterone receptors in the hippocampus of female rats

Authors: *L. M. BUYNACK[1], S. PATEL[2], E. GOMEZ-PERALES[3], J. LACASSE[4], W. G. BRAKE[5];
[1]CSBN, Concordia Univ., Montreal, QC, Canada; [2]Concordia Univ., Dollard-des-Ormeaux, QC, Canada; [3]Concordia Univ., Montreal, QC, Canada; [4]Concordia Univ., St. Catharines, ON, Canada; [5]Ctr. for Studies in Behav Neuro, Concordia Univ., Montreal, QC, Canada

Abstract: Sex hormones have been shown to influence a wide variety of memory systems. For example, ovarian hormones across the estrus cycle affect memory bias, favoring one memory system over another in the context of spatial navigation in either novel or familiar environments. Estrogens play a critical role in memory bias, with high levels of 17ß-estradiol (E2) promoting place memory, while low levels of E2 promote response memory. Progesterone in combination with E2 either 1 or 4 hours before testing, reverses the effects of high E2 and promotes bias toward response memory. While much is known about the role and mechanisms of estrogens, less research has focused on progesterone. To investigate the potential mechanisms by which progesterone may also be influencing memory bias, fluorescence immunohistochemistry was employed for nuclear progesterone receptors (nPR) and membrane progesterone receptors (mPR). nPR, mPRδ and mPRß were observed in brain tissue collected from ovariectomized female Long Evans rats under three hormonal conditions: low E2, high E2, and high E2 plus progesterone. Preliminary results show that mPRδ and mPRß are present in the hippocampus and entorhinal cortex. More specifically, mPRδ receptors are present in the CA3 region of the earliest anterior dorsal hippocampus, the hilus and stratum lacunosum-moleculare of the medial to posterior dorsal hippocampus, and the anterior lateral entorhinal cortex. mPRß receptors are present in the medial to posterior dorsal CA1, CA2, CA3, CA4, dentate gyrus and anterior lateral entorhinal cortex. Final results will include N=6 brains per hormone condition (N=18) and compare receptor expression across hormonal conditions throughout the hippocampus and entorhinal cortex. The presence of the membrane receptors suggest that progesterone may have both genomic and non-genomic effects in these areas.

Disclosures: L.M. Buynack: None. **S. Patel:** None. **E. Gomez-Perales:** None. **J. Lacasse:** None. **W.G. Brake:** None.

Poster

PSTR158. Molecular and Cellular Changes with Reproductive State and Hormones

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR158.11/HH2

Topic: F.02. Neuroendrocrine Processes and Behavior

Support: NRDIO grant K128278
Programme Széchenyi Plan Plus, RRF-2.3.1-21-2022-00011

Title: Cholinergic drive to GnRH neurons in mice

Authors: *Z. LIPOSITS[1], C. VASTAGH[1], I. FARKAS[1], V. CSILLAG[1], M. WATANABE[2], I. KALLO[1];
[1]Inst. of Exptl. Med., Budapest, Hungary; [2]Hokkaido Univ. Sch. Med., Sapporo, Japan

Abstract: Hypophysiotropic GnRH neurons orchestrate reproduction under control of neurotransmitters. Acetylcholine (ACh) has been shown to modify reproduction centrally, however, the exact target sites/s and the involved mechanisms haven't been clarified yet. To elucidate the putative networking between the central cholinergic and GnRH systems, studies were carried out in adult, male mice using various methods. 3DISCO immunocytochemistry revealed the innervation of cell bodies and dendrites of GnRH neurons by vesicular acetylcholine transporter (VAChT)-IR axons. The immunoelectron microscopic analysis identified cholinergic synaptic inputs to GnRH cells. Retrograde rabies virus labeling from GnRH-cre neurons explored the origin of the cholinergic afferents from the medial septum and diagonal band of Broca. Expression profiling and patch-clamp studies confirmed the expression of α4β2, α3β4 and α7 nicotine and M1-M5 muscarine ACh receptors (AChRs) in GnRH neurons. Carbachol (40 μM) first evoked an inward current, then decreased the frequency of mPSCs in these cells in the presence of TTX. The former action was prevented by mecamylamine (10 μM), the latter one by muscarine (10 μM) treatment confirming the involvement of both nicotine and muscarine AChRs in the regulation. ACh and carbachol also influenced the frequency of firing in GnRH neurons in a biphasic manner. For acceleration of firing nicotine and M1/M3 muscarine receptors, while for the inhibitory phase M2/M4 muscarine receptors were responsible. The actions of both facilitatory and inhibitory muscarine receptors were eliminated by tetrahydrolipstatin (THL) administration indicating the involvement of retrograde endocannabinoid signaling in the process. Optogenetic stimulation of channelrhodopsin-2 expressing cholinergic axons at 5 Hz exerted a biphasic effect on frequency of both firing and mPSCs in GnRH neurons similar to the pharmacological challenges. In a subpopulation of GnRH neurons (10%), the LED-evoked mPSCs at 0.2 Hz were abolished by picrotoxin, while at 5 Hz, in addition to picrotoxin, blockade of nicotine and muscarine receptors (by mecamylamine + atropine) was necessary for extinction of the effect. The finding implies that certain cholinergic afferents can co-transmit GABA with ACh in a frequency-dependent manner. In triple transgenic, Chat-Cre-Gq DREADD orchidectomized mice, clozapine-N-oxide (CNO) treatment evoked a significant, two-fold increase in both the basal and mean luteinizing hormone (LH) levels peaking at 30 minutes after ligand delivery. Collectively, the results indicate that ACh is a potent regulator of reproduction via hypophysiotropic GnRH neurons.

Disclosures: Z. Liposits: None. **C. Vastagh:** None. **I. Farkas:** None. **V. Csillag:** None. **M. Watanabe:** None. **I. Kallo:** None.

Poster

PSTR158. Molecular and Cellular Changes with Reproductive State and Hormones

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR158.12/HH3

Topic: F.02. Neuroendrocrine Processes and Behavior

Support: USDA NIFA Grant 2022-67015-37228

Title: Investigation into the role of estradiol in puberty onset of female sheep

Authors: *E. G. AERTS[1], A. C. THOMSON[1], M. J. GRIESGRABER[1], E. C. BOWDRIDGE[1], S. L. HARDY[1], R. L. GOODMAN[1], C. C. NESTOR[2], S. M. HILEMAN[1];
[1]West Virginia Univ., Morgantown, WV; [2]North Carolina State Univ., Raleigh, NC

Abstract: Puberty onset in many species is due to decreased inhibition by estradiol (E_2) of gonadotropin-releasing hormone (GnRH) and luteinizing hormone (LH) release. As GnRH neurons do not contain ERα, regulation by E_2 must be afferent to GnRH neurons. However, where in the hypothalamus E_2 acts is not completely understood. Two likely areas are the arcuate nucleus (ARC) and preoptic area (POA) of the hypothalamus because GnRH neurons are located primarily in the POA, and the ARC contains neurons that control GnRH secretion. Specifically, there is evidence in ewes supporting roles for ARC KNDy (containing kisspeptin, neurokinin B, and dynorphin), proopiomelanocortin (POMC,) and agouti-related peptide (AgRP) neurons in puberty onset. Our goal here was to determine if E_2 acts in the POA and/or ARC to regulate LH secretion prior to puberty. In addition, we investigated changes in ERα expression during pubertal development in ARC POMC and AgRP neurons; previous work had shown that ERα expression in KNDy neurons does not change during puberty. Two groups of ewes were ovariectomized and received guide tubes in either the POA (n=8) or the ARC (n=6). Two weeks later, blood samples were collected to assess LH secretion. Either E_2 or blank microimplants were placed and blood was collected a week later after which treatments and sampling was repeated in a crossover manner. Finally, microimplants were removed and subcutaneous (SQ) E_2 implants were inserted for one week and blood again collected. E_2 implants in the ARC reduced mean LH and LH pulse amplitude to that of means for SQ E_2-implanted ewes. In POA-implanted ewes, reduction in mean LH was similar to that seen in blank- and SQ-implanted ewes. No change in pulse frequency was noted for any treatment. In a second experiment, 3 groups of ewes were ovariectomized and received SQ E_2 implants at 5 (prepubertal; n=4), 8 (peripubertal, n=5), and 10 (postpubertal, n=5) months of age. Blood samples were collected two weeks later to assess LH secretion and hypothalamic tissue was collected and assessed for POMC, AgRP, and ERα via immunohistocyhemistry. Pulsatile LH release was least at 5 months, greatest at 10 months, and intermediate at 8 months. The percentage of ARC POMC or AgRP neurons that coexpressed ERα did not significantly change over development. These results indicate that E_2 acts more in the ARC than POA to regulate LH secretion prepubertally and that changes in POMC and AgRP ERα expression are likely not involved.

Disclosures: E.G. Aerts: None. **A.C. Thomson:** None. **M.J. Griesgraber:** None. **E.C. Bowdridge:** None. **S.L. Hardy:** None. **R.L. Goodman:** None. **C.C. Nestor:** None. **S.M. Hileman:** None.

Poster

PSTR158. Molecular and Cellular Changes with Reproductive State and Hormones

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR158.13/HH4

Topic: F.02. Neuroendrocrine Processes and Behavior

Support: JSPS KAKENHI 18H06037
JSPS KAKENHI 19K21172
JSPS KAKENHI 20K15650
JSPS KAKENHI 23K14064

Title: Vaginocervical stimulation activates AVPV Kisspeptin neurons

Authors: *S. NAKAMURA[1], A. HIGASHI[2];
[1]Nagoya Univ., Nagoya, Japan; [2]Okayama Univ. of Sci., Imabari, Japan

Abstract: The timing of copulation and ovulation need to coincide with each other to ensure fertilization in mammals. In this context, it is plausible that mating stimuli derived from male animals are involved in the neuroendocrine mechanisms that regulate gonadotropin-releasing hormone/luteinizing hormone surge and subsequent ovulation in females. In female rodents, *Kiss1* neurons located in the anteroventral periventricular nucleus (AVPV) are responsible for LH surge and ovulation. The present study aimed to investigate the effect of the copulatory stimulus, such as cervical stimulation, on the AVPV *Kiss1* neuronal activity in ovariectomized rats treated with a proestrous level of estradiol (OVX+E_2). OVX+E_2 rats cohabited with male rats for 1 hour from 17:00 h. Experimental groups were divided into two groups: the normal mating group (n = 4) and the mating without intromission group (n = 4) whose vagina was sealed to protect from penis insertion. Control animals (n = 4) did not cohabit with males. The rats were then perfused with 4% paraformaldehyde, and frozen sections with 50-µm were prepared for immunohistochemistry of c-Fos protein, a marker of neuronal activation, and *in situ* hybridization staining for the *Kiss1* gene. Penile intromission during copulation significantly increased the number of c-Fos-expressing *Kiss1* neurons in the AVPV ($p < 0.05$, one-way ANOVA followed by Holm test). The present study also examined whether artificial vaginocervical stimulation (VCS) with the sonic vibrator activates the AVPV *Kiss1* neurons in OVX+E_2 rats. The number of c-Fos-expressing AVPV *Kiss1* neurons was significantly higher in the VCS group (n = 4) than in the control group (n = 4) ($p < 0.05$, Student's *t*-test). Finally, the effect of VCS on the number of oocytes ovulated was investigated in intact female rats. There was no significant difference in the number of ovulated oocytes after VCS at 13:00 or at 17:00 h at proestrus in female rats. The present study suggests that AVPV *Kiss1* neurons are one of the targets of signals from the vaginocervix of copulatory stimulation in female rats. This mechanism may contribute to ensuring ovulation when female rats receive cervical stimulation during copulation.

Disclosures: S. Nakamura: None. **A. Higashi:** None.

Poster

PSTR158. Molecular and Cellular Changes with Reproductive State and Hormones

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR158.14/HH5

Topic: F.02. Neuroendrocrine Processes and Behavior

Support: Undergraduate Research Fund, 2023

Title: B2 SINE RNA is a novel regulator of androgen receptor transcriptional activity

Authors: *T. A. RICHTER[1], R. G. HUNTER[2];
[1]Univ. of Massachusetts, Boston Dept. of Psychology, Boston, MA; [2]Univ. of Massachusetts Boston, Univ. of Massachusetts, Boston, MA

Abstract: The glucocorticoid receptor plays a role in the pathophysiology of some stress-related psychological disorders like post-traumatic stress disorder (PTSD), schizophrenia, and major depressive disorder (MDD) to name a few. Some of these disorders display a sex difference in their prevalence or symptomatology, while genome-wide association studies fail to implicate protein-coding genes as responsible for this sex difference. The non-coding genome represents a possible area of interest to explain this difference. B2 SINE RNA is derived from a transposable element, regulated by glucocorticoids, and in turn, regulates the transcriptional activity of glucocorticoid receptor. Currently, it is unknown whether this non-coding RNA regulates the transcriptional activity of androgen receptor (AR) which are found throughout the brain and body. Here, we describe how B2 SINE RNA regulates androgen receptor transcriptional activity. We discovered a *de novo* motif within B2 SINE RNA *in silico* that aligned with both the general hormone response element (HRE) consensus sequence ($p<0.001$) and the androgen receptor specific response element (ARE) consensus sequence ($p<0.05$). Furthermore, we used chromatin immunoprecipitation (ChIP) in lymph node carcinoma of the prostate cells (LNCaP) to confirm the transcriptional activity change of AR. This cell line is commonly used to study the full-length androgen receptor *in vitro*. We transfected this cell line with either full-length B2 SINE RNA derived from a gene block or a scrambled version of this RNA sequence. RT-qPCR was utilized to quantify the change of expression of common androgen-regulated genes in the presence or absence of this RNA such as FKBP5, PSA, OPRK1, NKX3-1, and TMPRSS2. These results, similar to what we have previously reported with the glucocorticoid receptor *(*Bartlett *et al.*, 2023), show that there exists a specific recognizable sequence within the RNA of B2 SINE that androgen receptor can recognize. Binding of AR to this RNA will then change the expression of androgen-related genes. Since B2 SINE RNA expression is regulated by glucocorticoids, this could serve as an explanation for the sex differences observed in some stress-related neuropsychological disorders. Additionally, this work begins to establish B2 as a novel translational target for the treatment of disorders involving androgen receptor signaling.

Disclosures: T.A. Richter: None. **R.G. Hunter:** None.

Poster

PSTR158. Molecular and Cellular Changes with Reproductive State and Hormones

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR158.15/HH6

Topic: F.02. Neuroendrocrine Processes and Behavior

Title: Molecular mechanisms underlying the regulation of metabolic function by ovarian hormones

Authors: *L. OULDIBBAT, D. ROCKS, B. SAMPSON, M. KUNDAKOVIC;
Biol. Sci., Fordham Univ., Bronx, NY

Abstract: Throughout the female reproductive period, ovarian hormone estradiol plays a protective role in regulating different domains of metabolic function including body weight, appetite, and energy expenditure. Females are at increased risk for metabolic dysfunction especially during the menopausal transition as declines in estrogen lead to decreased activity and increased fat mass. In the functionally diverse hypothalamus, estrogen targets distinct nuclei populations to maintain energy balance. However, little is known about the molecular mechanisms through which ovarian hormones regulate these specific cellular groups. To address this question, we first examined metabolic phenotypes of ovariectomized (OVX) and ovary-intact (cycling) C57BL/6 female mice across the estrous cycle. Animals were monitored from 9-21 weeks to track weight, feeding, activity, and adiposity. We find that OVX animals demonstrate up to 40% weight increase and up to 100% fat mass increase compared to cycling animals. Upon examining weights across the estrous cycle, we find a drop in weight in the estrus phase following decreased feeding observed in the high-estrogenic, proestrus phase. On the contrary, there was a consistent weight increase in OVX animals across time, due to increased feeding and decreased activity. To address the underlying molecular mechanisms, we analyzed the expression of estrogen-sensitive metabolic genes in the bulk hypothalamus and white adipose tissue at 21 weeks using qRT-PCR. In the hypothalamus, estrogen receptor α (Esr1) expression varied with the estrous cycle with highest expression in proestrus and lowest expression in OVX mice. Pro-opiomelanocortin receptor (POMC), an anorexigenic neuropeptide contributing to negative energy balance, demonstrated highest expression in proestrus and lowest expression in OVX mice. Agouti-related protein (AgRP), an orexigenic neuropeptide, demonstrated higher expression in diestrus consistent with lower estradiol levels. In the adipose tissue, OVX animals had significantly more Leptin receptor (Lepr) expression, consistent with excess proliferation of adipocytes. Ghrelin receptor (Ghsr) exhibited increased expression in OVX mice consistent with their increased feeding. We are currently performing single-cell analysis of the hypothalamus across the estrous cycle and in OVX animals to identify cellular populations and molecular drivers that are sensitive to ovarian hormone levels. These studies will provide the link between the metabolic phenotype and ovarian hormone regulated molecular mechanisms in the hypothalamus, providing novel opportunities for treatment of metabolic disorders.

Disclosures: L. Ouldibbat: None. **D. Rocks:** None. **B. Sampson:** None. **M. Kundakovic:** None.

Poster

PSTR158. Molecular and Cellular Changes with Reproductive State and Hormones

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR158.16/HH7

Topic: F.02. Neuroendrocrine Processes and Behavior

Title: Estrogen-responsive MC4R^{+}neurons in the ventrolateral ventromedial hypothalamic nucleus VMHvl project to arousal centers of the pons

Authors: *L. E. CABRERA-ZAPATA[1], W. C. KRAUSE[1], X. DUAN[2], H. A. INGRAHAM[1]; [1]Dept. of Cell. and Mol. Pharmacol., [2]Dept. of Ophthalmology, Univ. of California San Francisco, San Francisco, CA

Abstract: The medial basal hypothalamus (MBH) contains sexually dimorphic clusters of neurons that regulate distinct sex-specific physiology and behaviors. In females, the preovulatory surge of estrogen engages the hypothalamus to temporarily increase energy expenditure, enabling it to coordinate peak sexual receptivity with increased physical activity, thus maximizing reproductive success. Our lab has recently identified a small subset of estrogen-sensitive, melanocortin-4 receptor (*Mc4r*)-expressing neurons in the ventrolateral ventromedial hypothalamic nucleus (VMHvl) that control this spontaneous locomotive behavior. VMHvl^{MC4R} neurons are highly enriched for the estrogen receptor alpha (ERα). Subsequent upregulation of *Mc4r* expression by ERα in these VMHvl neurons leads to increased spontaneous activity. Here we focused on the identification and characterization of the VMHvl^{MC4R} neuronal projections into specific nuclei of the pons, a hindbrain region mediating a wide variety of homeostatic behaviors and autonomic functions ranging from sexual and locomotor arousal, appetite, thermoregulation, micturition, and breathing. Male and female adult *Mc4r-t2a-cre* mice were stereotaxically injected in the VMHvl with a Cre-dependent anterograde trans-synaptic tracer (AAV2-CAG-DIO-mWGA-mCherry-WPRE). Four to five weeks post-injection, brain tissue was fixed, cryosectioned, and immunolabeled for imaging and mapping the VMHvl^{MC4R} neuronal projections and post-synaptic pontine neurons. We found post-synaptic expression of the tracer in pontine neuronal populations, with enrichment of synaptic input into the locus coeruleus (LC) and Barrington's nuclei. Remarkably, these mCherry-marked neuronal clusters showed significant ERα expression levels. We also localized ERα staining to clusters of neurons in the rostro-lateral and cauda-dorsal portions of the parabrachial, including the Kölliker-Fuse nucleus. Having mapped the VMHvl^{MC4R}-pons neuronal circuit, we are currently identifying which neuronal subset within the LC connects with the VMHvl^{MC4R} and asking how estrogen signaling in these different clusters affects physiological and behavioral outputs, such as energy expenditure, physical activity, and arousal in male and female mice.

Disclosures: L.E. Cabrera-Zapata: None. **W.C. Krause:** None. **X. Duan:** None. **H.A. Ingraham:** None.

Poster

PSTR158. Molecular and Cellular Changes with Reproductive State and Hormones

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR158.17/HH8

Topic: F.02. Neuroendrocrine Processes and Behavior

Title: Uterus Weights as Proxy for Estrogen Levels in POMC-Deficient Mice

Authors: *B. MCHOES[1,2], R. RICKS[2], Z. THOMPSON[2];
[1]Utah Valley Univ., Springville, UT; [2]Utah Valley Univ., Orem, UT

Abstract: Expression of the pro-opiomelanocortin (POMC) gene in the hypothalamus leads to the production of several hormones, including α-melanocyte stimulating hormone (α-MSH) and β-endorphin (β-EP). α-MSH is a metabolic response hormone that helps to regulate food intake in response to energy needs. In humans, mutations in the POMC gene lead to reduced expression of the gene, and to excessive hunger and eventually obesity. We are studying a mouse model with a recessive mutation that results in little to no expression of the POMC gene. Like POMC-deficient humans, these mice exhibit hyperphagia, early-onset obesity and they also are infertile. We are attempting to elucidate the cause of this infertility through several projects. Estrogen levels have a powerful effect on fertility in mammals. One way to measure estrogen levels by proxy is via uterus weight, which increases with increased levels of estrogen. The goal of this project is to understand the effect of decreased expression of the POMC gene on estrogen levels via measuring the weight of the uterus. After performing a hysterectomy, the uterus and ovaries are weighed by a researcher blind to genotype, and this data is used as an indicator of estrogen in the mouse. As a secondary factor, the stage of the estrus cycle the mouse was in at the time of the hysterectomy is recorded. Preliminary analyses indicate that female POMC-deficient mice have a lower uterus weight than female wild-type mice, though we are collecting additional data to more fully answer this question. As we collect more data on estrogen levels and infertility in POMC-deficient mice, we hope to better understand the effects POMC deficiency has on humans as well.

Disclosures: B. McHoes: None. **R. Ricks:** None. **Z. Thompson:** None.

Poster

PSTR158. Molecular and Cellular Changes with Reproductive State and Hormones

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR158.18/HH9

Topic: F.02. Neuroendrocrine Processes and Behavior

Support: JSPS KAKENHI Grant Number 22H00397

Title: Local administration of neurokinin B at the arcuate nucleus of goats accelerates the activity of the gonadotropin-releasing hormone pulse generator

Authors: *S. OHKURA[1], T. YAMAMURA[2], S. NAKAMURA[1], Y. WAKABAYASHI[2]; [1]Nagoya Univ., Nagoya, Japan; [2]Inst. of Livestock and Grassland Sci., Natl. Agr. and Food Res. Organization, Tsukuba, Japan

Abstract: Kisspeptin neurons in the arcuate nucleus (ARC), which co-express neurokinin B (NKB) and dynorphin A, are called KNDy neurons. These neurons are candidates for the intrinsic source of the gonadotropin-releasing hormone (GnRH) pulse generator. The cerebroventricular and intravenous administration of NKB or its receptor (NK3R) agonist induces the characteristic increase in the multiple unit activity (MUA volley) in the ARC, an electrophysiological manifestation of the GnRH pulse generator activity, and stimulates the subsequent pulsatile GnRH/luteinizing hormone (LH) secretion. However, the mechanism responsible for the neural activation of the GnRH pulse generator is unclear. The present study aims to test the hypothesis that NKB acts on the KNDy neurons directory and that the signal is transmitted bilaterally to a population of KNDy neurons in the ARC using the electrophysiological and histochemical technique in goats. Bilateral electrodes aimed at a cluster of KNDy neurons were inserted into the ARC of ovariectomized goats for MUA recording. The unilateral administration of NKB or vehicle in the close vicinity of KNDy neurons under simultaneous MUA recording from both sides revealed that only NKB evoked the MUA volley immediately after administration. The timing of the MUA volley evoked on the ipsilateral side was synchronized to that on the contralateral side. The double-labeled *in situ* hybridization for *KISS1* and *TACR3*, which encode kisspeptin and NK3R, revealed that most KNDy neurons co-expressed *TACR3* (96.2 ± 0.2%). Tract tracing histochemistry using biotinylated dextran amine (BDA), an anterograde tracer, indicated that axons projecting from NKB neurons in the ARC were directly apposed to other NKB neuronal cells located bilaterally in the ARC, indicating that KNDy neurons are bilaterally interconnected in the ARC via NKB-containing fibers. These results suggest that NKB administered locally into the ARC directly stimulates KNDy neurons, following which the stimulatory signal is immediately transmitted to the entire population of KNDy neurons on both sides of the ARC via connection with their fibers. This mechanism might play a critical role in synchronizing bursting activity among KNDy neurons, thereby generating neural signals of the GnRH pulse generator that govern pulsatile GnRH secretion.

Disclosures: S. Ohkura: None. **T. Yamamura:** None. **S. Nakamura:** None. **Y. Wakabayashi:** None.

Poster

PSTR158. Molecular and Cellular Changes with Reproductive State and Hormones

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR158.19/HH10

Topic: F.02. Neuroendrocrine Processes and Behavior

Support: NIH-R15MH118692

Title: Dihydrotestosterone regulation of stress-related behaviors in mice exposed to subchronic variable stress

Authors: *K. A. RYBKA, J. J. LAFRICAN, K. E. PARRA, D. G. ZULOAGA;
Psychology, Univ. At Albany State Univ. of New York, Albany, NY

Abstract: Androgen actions through androgen and estrogen receptors have been shown to regulate stress-related behaviors as well as hypothalamic-pituitary-adrenal (HPA) axis responsiveness. However, the role of androgens and the androgen receptor in regulating stress-related behaviors and the HPA axis under chronic stress conditions are unclear. Here we utilized a subchronic variable stress (SCVS) paradigm to evaluate the role of androgens and the androgen receptor in mediating behavioral changes after repeated stress. Wild type C57BL6/J adult male mice were gonadectomized and given either a blank pellet or a pellet containing dihydrotestosterone (DHT), which preferentially binds the androgen receptor. Half of the animals of each group were either non-stressed controls (Control Blank or Control DHT) or stressed (Stress Blank or Stress DHT). In brief, the SCVS paradigm consisted of three days of an hour-long stressor that alternated every day and was repeated in the same order for three more days. Stressors included footshock, tail suspension, and restraint stress. Behavior testing to evaluate stress-related behaviors consisted of the splash test, sucrose preference, novelty suppressed feeding (NSF), open field test (OFT), and forced swim test (FST). In the OFT, unstressed DHT-treated mice showed a greater number of center entries when compared to mice that received a blank pellet, indicating decreased anxiety-like behaviors. However, the stress-reducing effect of DHT was not observed in mice exposed to SCVS. In the FST, unstressed mice receiving DHT spent less time floating compared to unstressed mice that received a blank pellet, indicating a more active coping pattern in DHT mice which could indicate a protective mechanism of androgen receptor in despair-like behavior. However, again this protective effect of DHT was not observed in mice exposed to SCVS. No significant differences were found in the splash test, sucrose preference, or NSF. Analysis of corticosterone (CORT) in mice exposed to the FST revealed that overall DHT reduced CORT levels but no difference in CORT was found between the unstressed and SCVS groups. Preliminary results suggest that androgen actions through androgen receptors reduce behavioral stress responses, although these beneficial effects may be lost under conditions of repeated stress.

Disclosures: **K.A. Rybka:** None. **J.J. Lafrican:** None. **K.E. Parra:** None. **D.G. Zuloaga:** None.

Poster

PSTR158. Molecular and Cellular Changes with Reproductive State and Hormones

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR158.20/HH11

Topic: F.02. Neuroendrocrine Processes and Behavior

Support: R01AG070072

Title: Advanced infrared image analysis for monkey hot flush detection: a deep learning approach

Authors: *I. MERCHENTHALER[1,3], H. HOORFAR[1], M. LANE[1], A. PUCHE[2];
[1]Epidemiology & Publ. Hlth., [2]Anat. and Neurobio., Univ. of Maryland, Baltimore, MD;
[3]Oregon Natl. Primate Res. Ctr., Beaverton, OR, OR

Abstract: Advanced Infrared Image Analysis for Monkey Hot Flush Detection: A Deep Learning ApproachAuthors: *H, HOORFAR[1], M. LANE[1], H.F. URBANSKI[2], A. C. PUCHE[1], I. MERCHENTHALER[1], [1]University of Maryland Baltimore; [2]Oregon National Primate Research Center
Disclosures: H. Hoorfar: none; **M. Lane**: none; **H.F. Urbanski**, none; **A.C. Puche:** none; **I. Merchenthaler:** none.
Menopause is an inevitable stage in normal human aging, affecting the quality of life of millions of individuals. The most 'unbearable' symptoms are hot flushes (HFs). Among current treatments of HFs, only estrogen (E2) therapy has satisfactory efficacy, but it has significant side effects by stimulating uterine and breast cell proliferation. Although there have been extensive efforts to develop novel therapies, the lack of animal model(s) has had a negative impact on success of this effort.In this study we developed an old-world highly-translational animal model that undergoes a menopausal process identical to that seen clinically in individuals with a uterus as well as processes to detect HFs using non-invasive thermal imaging. Imaging data is autonomously analyzed using Convolutional Neural Network (CNN) models programmed in the Python programming language. The result is an efficient CNN algorithm that can detect primate thermal facial features and automated HF detection, with a computational load allowing analysis to parallel image acquisition (i.e., 24 hours of imaging data takes less than 25 minutes to process). The entire image processing pipeline operates automatically using deep learning techniques. The algorithm has been trained extensively to effectively consider various parameters of an image, including body position, lens distortions, and facial features, to accurately determine the temperature of face. By analyzing temperature variations across different parts of the faces, our algorithm can accurately detect and identify key facial features which play crucial roles in heat dissipation with negligible false positive/false negative HFs.This research relies on data analysis due to the millions of infrared images collected by 24/7 video imaging of the subjects. Through this comprehensive analysis, we will be able to provide novel observations on the occurrence and patterns of HFs in monkeys, shedding light on potential underlying factors and implications. Our approach demonstrates the efficacy of deep learning and CNNs in autonomous systems extracting meaningful information from complex datasets, providing a solid foundation for further research in this field. This approach will help with the evaluation of novel strategies aimed at preventing HFs.Supported by R01AG070072

Disclosures: I. Merchenthaler: None. **H. Hoorfar:** None. **M. Lane:** None. **A. Puche:** None.

Poster

PSTR159. Early-Life Stress: Molecular Mechanisms and Cellular Effects

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR159.01/HH12

Topic: F.03. Stress and the Brain

Support: Research Sector, Kuwait University: Grant No. YM04/21

Title: Interleukin 6 contributes to the lasting impact of prenatal immune challenge on GABAergic inhibitory markers within the prefrontal cortex_A sex-dependent effect.

Authors: *A. MOUIHATE, R. ALHARBI;
Physiol., Col. of Medicine, Kuwait Univ., Kuwait City, Kuwait

Abstract: Experimental evidence suggests that maternal immune activation (MIA) with the bacterial active ingredient lipopolysaccharide (LPS) leads to a reduced GABAergic inhibitory tone in the offspring's prefrontal cortex (PFC). Because interleukin-6 (IL-6) mediates the long-lasting behavioral and cognitive dysfunctions induced by MIA, we sought to assess whether IL-6 contributes to the MIA effects on the cell density of GABAergic neurons, the inhibitory synapses, and the expression levels of chloride transporters NKCC1 and KCC2 in the prefrontal cortex of 30 days old male and female rat offspring. Pregnant rats were given intraperitoneal injections of either pyrogen-free saline or lipopolysaccharide (LPS, 100 µg/Kg) in the presence or the absence of an IL-6 neutralizing antibody (IL-6Ab, 10 µg/Kg), on gestation days (GD)15, GD17 and GD19. Each rat group (Saline-IgG, LPS-IgG, Saline-IL-6Ab, LPS-IL6Ab) consists of 5-6 pregnant rats. The density of GABAergic interneurons (parvalbumin, somatostatin, 5HT3A types) and the density of inhibitory synapses (the juxtaposition of gephyrin and VGAT) were monitored in the prefrontal cortex of 30 days old male and female rat offspring using fluorescent immunohistochemistry. The expression levels of NKCC1 and KCC2 protein were monitored using Western blot. MIA reduced the cell density of parvalbumin- and 5HT3A-containing interneurons and the density of inhibitory synapses in the prefrontal cortex of female but not male rat offspring. MIA reduced the protein expression levels of NKCC1 but not that of KCC2 in both male and female rat offspring. These effects were not seen when the IL-6Ab was prenatally co-administered with LPS. These data suggest that female rats are more prone to MIA-induced reduction in the inhibitory system within the PFC. These effects were largely mediated by the LPS-activated IL-6. This study highlights the importance of exploring the early brain development and plasticity that might underlie a sex-dependent behavioral dysfunction later in life.

Disclosures: A. Mouihate: None. **R. AlHarbi:** None.

Poster

PSTR159. Early-Life Stress: Molecular Mechanisms and Cellular Effects

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR159.02/HH13

Topic: F.03. Stress and the Brain

Support: NIH Grant MH73136
NIH Grant MH096889
The Bren Foundation

Title: Defining the functional roles of GABA and CRH in the CRH/GABA BLA to NAc pathway

Authors: *M. BIRNIE[1], M. HARDY[1], G. B. DE CARVALHO[1], L. TANIGUCHI[1], B. G. GUNN[1], L. Y. CHEN[1], Y. CHEN[1], N. J. JUSTICE[2], T. BARAM[1];
[1]Univ. of California-Irvine, Irvine, CA; [2]Ctr. for Metabolic and Degenerative Dis., Institue of Mol. Medicine, Univ. of Texas Hlth. Sci. Ctr., Houston, TX

Abstract: Background: The nucleus accumbens (NAc) is a major component of the reward circuit and key structure mediating pleasure, motivation, and emotional processes. Multiple inputs converge onto the NAc to modulate reward behaviors, including the basolateral amygdala (BLA). We recently reported a GABAergic projection from the BLA to the NAc that expresses the stress-sensitive neuropeptide corticotropin-releasing hormone (CRH). Specifically, this projection suppressed reward behavior. Here, we sought to identify the individual and collective contributions of CRH and GABA mediated by this projection during reward behavior. Methods: Pairing viral-genetic approaches with CRH-ires-Cre mice and Cre-dependent viruses, we previously identified CRH/GABA+ BLA projections to the NAc. To determine the contributions of CRH and GABA to reward behavior, we crossed CRH-ires-Cre with CRFR1-Flp mice and utilized chemogenetic, optogenetic and electrophysiological strategies in male and female mice. Using slice physiology, an excitatory cre-dependent optogenetic (AAV1-DIO-ChR2) virus was injected into the BLA, followed by light stimulation and CRH receptor 1 and/or 2 antagonism in the NAc. In freely behaving mice, the excitatory cre-dependent chemogenetic (AAV2-DIO-hM3Dq) virus was injected into BLA, followed by medial NAc shell targeted microinjections of CNO (with and without CRH receptor 1/2 antagonism). In behavior, the function of CRH was tested using reward, and non-reward related tasks. Results: Viral-genetic tracing combined with slice electrophysiology identified a CRH/GABA projection from the BLA to the medial NAc shell that was reliably inhibited with the $GABA_AR$ antagonist, picrotoxin. In freely behaving mice, stimulating this projection using DREADDs suppressed reward behavior. Pharmacological experiments combining CRH-ires-Cre mice, inhibitory DREADDs and CRH receptor 1 (NBI 30775) and 2 (ASVG30) antagonists to delineate the actions of CRH and GABA in reward behavior are in progress. Experiments crossing CRH-ires-Cre mice with CRFR1-Flp mice to conditionally knockout CRFR1 using CRISPR/Cas9 technologies, whilst allowing for the excitation of the CRH+ BLA-NAc projection during slice electrophysiology and in vivo behavior experiments are underway. Conclusions: Here, we delineate the roles of CRH and GABA on the recently described CRH/GABA BLA-NAc projection and establish their role in mediating the effects of stress on reward behavior. These discoveries may provide potential selective targets for prevention and intervention in several mental illnesses including depression and substance use disorder.

Disclosures: M. Birnie: None. **M. Hardy:** None. **G.B. De Carvalho:** None. **L. Taniguchi:** None. **B.G. Gunn:** None. **L.Y. Chen:** None. **Y. Chen:** None. **N.J. Justice:** None. **T. Baram:** None.

Poster

PSTR159. Early-Life Stress: Molecular Mechanisms and Cellular Effects

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR159.03/HH14

Topic: F.03. Stress and the Brain

Support: George E. Hewitt Foundation for Medical Research Postdoctoral Fellowship
MH 096889

Title: A double TRAP reveals the influence of early life adversity on the translatome of the paraventricular nucleus of the thalamus (PVT)

Authors: *A. FLORIOU-SERVOU[1,2], H. Y. LIANG[3], K. KIM[4], M. GANTUZ[3], G. D. ANGELES[2], R. ROBERTS[2], A. MORTAZAVI[3], T. Z. BARAM[2,5];
[2]Dept. of Anat. & Neurobio., [3]Dept. of Developmental & Cell Biol., [4]Dept. of Informatics, [5]Dept. of Pediatrics, [1]UC Irvine, Irvine, CA

Abstract: Background: Early-life adversity (ELA) is associated with cognitive and mental health problems later in life. Evidence in rodents points to a causal role of ELA, with structural and functional changes in the brain's reward circuitry. However, the mechanisms through which ELA might change the maturation and function of the brain's reward circuit remain poorly understood. One emerging key node of the reward circuit is the paraventricular nucleus of the thalamus (PVT). Most importantly, according to data from our lab, the PVT is strongly and almost exclusively activated in the mouse brain early in life. This raises the possibility that the PVT encodes the ELA experience and influences reward-seeking behaviors later in life. Here, we explore how the PVT is itself affected by ELA. To this end, we explore the translatome of PVT cells activated early in life, and test the hypothesis that ELA causes enduring changes in their translational profiles, both at rest and in the context of reward. **Methods**: We crossed two strains of mice: a driver line expressing Fos-dependent Cre^{ERT2} that allows activity-dependent genetic labeling (TRAP2), with mice expressing a Cre-dependent ribosomal tag allowing translating ribosome affinity purification (TRAP). This approach isolated actively translated mRNA from cells that are activated during P6-P8, when mice are raised in either typical (control group) or ELA conditions. We collected the midline thalamus containing the whole PVT from 2-3 months old mice in baseline conditions, or one hour after the start of a reward paradigm. Subsequently we isolated the RNA bound to tagged ribosomes and performed next generation RNA sequencing. Mice from different groups were randomized during tissue collection and processing, and the experimenter was blinded throughout the experiment. **Results**: A. Strong enrichment in genes that are highly expressed in the PVT such as *Snca* and *Calb2*, and a reduction in genes that are less expressed in the PVT such as *Slc17a7* and *Fras1*, confirmed that the majority of the isolated RNA was from PVT cells. B. Relatively few genes were differentially expressed in adult ELA vs control mice during baseline conditions. C. In contrast, exposure to reward induced very different translatomic responses in ELA vs control females. **Conclusions**: Our results indicate that ELA primes the PVT, influencing the way that the female PVT responds to reward. **Funding**: This work was supported by the George E. Hewitt

Foundation for Medical Research Postdoctoral Fellowship and by the National Institutes of Health Grant MH096889.

Disclosures: **A. Floriou-Servou:** None. **H.Y. Liang:** None. **K. Kim:** None. **M. Gantuz:** None. **G.D. Angeles:** None. **R. Roberts:** None. **A. Mortazavi:** None. **T.Z. Baram:** None.

Poster

PSTR159. Early-Life Stress: Molecular Mechanisms and Cellular Effects

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR159.04/HH16

Topic: F.03. Stress and the Brain

Support: NIH R00MH115096
NIH R01MH129643
PNI Research Innovator Award
New York Stem Cell Foundation

Title: Effects of overexpression of Otx2 on thyroid hormone signaling

Authors: *S. BENNETT[1], J.-A. BALOUEK[2], C. J. PENA[3];
[2]Princeton Univ., [1]Princeton Univ., Princeton, NJ; [3]Princeton Univ., Princeton Neurosci. Inst., Princeton, NJ

Abstract: Thyroid hormones play a major role in the developing brain influencing processes such as dopamine neuron differentiation and formation. We have previously demonstrated early life stress (ELS) transiently suppressed a transcription factor, orthodenticle homeobox (Otx2) in dopamine neurons, which is also known to be regulated by thyroid hormone signaling. Here we sought to understand whether thyroid hormone indeed mediated the impact of ELS on Otx2 in ventral tegmental area (VTA) dopamine neurons, and on behavior. First, we find that ELS increases thyroid stimulating hormone in the juvenile period, indicative of suppressed juvenile thyroid function. We hypothesized that Otx2 was downstream of these changes. To test this hypothesis, we mimicked ELS by suppressing thyroid function via antithyroid agent, propylthiouracil (PTU) administered to animals through chow, and overexpressed Otx2 in VTA. Moreover, we tested this 2x2 manipulation in two different time windows to assess whether there are sensitive periods for the effects of thyroid inhibition and Otx2: a juvenile (P15-P17) time point, or an adult (P70-P75) time point. Animals were injected bilaterally in the VTA with HSV-Otx2 and underwent adult social defeat stress conditions. Two measures of behavior, social interaction and open field test, were performed before and after adult stress in order to test both baseline behavior and stress sensitivity. We found a significant effect of PTU on baseline open field behavior in both adult and juvenile conditions. Finally, we are performing qPCR to quantify expression of genes associated with thyroid or dopamine function to determine whether Otx2 rescue also rescued the effect of thyroid suppression at a molecular level. Based on these and

previous studies, thyroid hormones may bridge between the experience of ELS and downstream transcriptional changes to mediate stress-responsive behavior.

Disclosures: **S. Bennett:** None. **J. Balouek:** None. **C.J. Pena:** None.

Poster

PSTR159. Early-Life Stress: Molecular Mechanisms and Cellular Effects

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR159.05/HH17

Topic: F.03. Stress and the Brain

Support: Brain and Behavior Research Foundation NARSAD
NIH R00MH115096
NIH R01MH129643
Princeton Neuroscience Institute Research Innovator Award
New York Stem Cell Foundation
Robin Chemers Neustein

Title: Transcriptional signatures of early-life stress and antidepressant treatment efficacy

Authors: *S. T. PAREL, C. J. CHENG, C. J. PEÑA;
Princeton Neurosci. Inst., Princeton Univ., Princeton, NJ

Abstract: Early-life stress (ELS) is a major risk factor for depression and may also lead to poor response to first-line antidepressant treatments in the future. However, the effect of ELS on the efficacy of antidepressant treatments remains unclear. In the brain, the nucleus accumbens (NAc) has been shown to be important in depression and response to antidepressant drugs. In the mouse NAc in particular, ELS may sensitize individuals to future stress, and these changes can be detected at the molecular resolution of gene expression. To test our hypothesis that ELS produces lasting transcriptional changes in the NAc that predict non-response to antidepressant treatment, we integrated cross-species transcriptomic analyses on three independent RNA-sequencing datasets: 1) a mouse model for ELS and/or adult stress; 2) antidepressant imipramine or ketamine treatment response or failure in a mouse model of adult stress; and 3) antidepressant escitalopram or desvenlafaxine treatment response or failure in patients with major depression. The mouse datasets include samples from the NAc, and the human dataset represents samples from patient blood. We recently found that NAc transcriptomic signatures of ELS prior to adult stress can predict altered transcriptomic patterns associated with treatment non-response across species and multiple classes of antidepressants. To complement these previous analyses that were based on differential gene expression, we sought to understand how gene network connectivity may be altered by stress and related to antidepressant response in our current work. We constructed gene co-expression networks to identify hub genes that may be key drivers of wider transcriptional changes common to both ELS and antidepressant outcomes. We are then testing through quantitative PCR how these candidate hub genes may indeed be altered in the

mouse NAc and behavior after stress experience and pharmacological treatment with antidepressants. These studies provide neurobiological evidence for molecular adaptations in the brain related to early-life stress that contribute to antidepressant treatment response.

Disclosures: S.T. Parel: None. **C.J. Cheng:** None. **C.J. Peña:** None.

Poster

PSTR159. Early-Life Stress: Molecular Mechanisms and Cellular Effects

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR159.06/HH18

Topic: F.03. Stress and the Brain

Support: India Alliance Early career fellowship IA/E/18/1/504310
Sri Ramakrishna Paramhansa Grant 2019

Title: Early life adversity perturbs mitostasis and drives inflammaging in the rat hippocampus

Authors: *P. R. CHAUDHARI[1], A. SINGLA[1], K. KUKKEMANE[1], S. SURYAVANSHI[1], S. E. FANIBUNDA[1,3], P. TIWARI[1], V. DEWAN[1], A. PRADHAN[1], S. MENDON[1], U. KOLTHUR-SEETHARAM[1,2], A. D. B. VAIDYA[3], C. SANDI[4], V. A. VAIDYA[1];
[1]Dept. of Biol. Sci., Tata Inst. of Fundamental Res., Mumbai, India; [2]Tata Inst. of Fundamental Res., Hyderabad, India; [3]Kasturba Hlth. Society, Med. Res. Ctr., Mumbai, India; [4]Brain Mind Institute, Ecole Polytechnique Federale de Lausanne, Lausanne, Switzerland

Abstract: Early life environment shapes the development of neurocircuits involved in modulation of emotional behavior. Adverse experiences during early life are known to program enhanced anxio-depressive behaviors in adulthood, and to evoke perturbed neuroendocrine and behavioural responses to stress. Early stress is also linked to accelerated aging-related changes including, dendritic atrophy, decreased hippocampal neurogenesis, reduced neuronal survival and enhanced cognitive decline. However, the molecular and cellular changes evoked by early adversity that contribute to these phenotype are yet to be fully delineated. Aging is linked to mitochondrial dysfunction and inflammation, and we hypothesized that early adversity may disrupt these processes thus impact the nature of aging. Using a rodent model of early adversity, maternal separation (MS) which is reported to enhance adult anxiety and depressive-like behaviour, accelerate aging, and perturb neuroendocrine stress responses that endure across the life span, we addressed whether this was accompanied by perturbed mitostasis and peripheral-central inflammatory changes. We provide novel information that a history of MS is associated with a robust dysregulation in mitostasis, with a decline noted in mitochondrial biogenesis and enhanced mitophagy, accompanied by an inflammatory state evoked in the hippocampus, which emerges in an age-dependent manner. The changes in the brain are also associated with robust peripheral inflammatory changes, which appear to precede the neuroinflammation. This is accompanied by impaired mitochondrial function and morphology, as well as cognitive decline in middle-aged rats with a history of MS. Concomitantly, we also note a compromise of the

blood brain barrier in the hippocampus of MS rats as compared to age-matched control rats which emerges correlated with the onset of central inflammation. Studies are under way to assess the influence of metabolic interventional strategies that may serve to reverse the mitochondrial and inflammatory changes that we observe following early adversity.

Disclosures: **P.R. Chaudhari:** None. **A. Singla:** None. **K. Kukkemane:** None. **S. Suryavanshi:** None. **S.E. Fanibunda:** F. Consulting Fees (e.g., advisory boards); S.E.F. serves as a consultant to Beckley Psytech which is not relevant to the current work. **P. Tiwari:** None. **V. Dewan:** None. **A. Pradhan:** None. **S. Mendon:** None. **U. Kolthur-Seetharam:** None. **A.D.B. Vaidya:** None. **C. Sandi:** None. **V.A. Vaidya:** None.

Poster

PSTR159. Early-Life Stress: Molecular Mechanisms and Cellular Effects

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR159.07/HH19

Topic: F.03. Stress and the Brain

Support: NIH NIGMS P20GM103423

Title: Early life adversity leads to changes in DNA methylation in parvalbumin cells: possible age- and sex-specific epigenetic driver of vulnerability in rats

Authors: E. S. NOEL[1], Y. PEÑA[1], S. M. BONAUTO[1], A. CHEN[1], S. ELLIS[1], A. HOPKINS[3], S. MILLER[3], ***J. A. HONEYCUTT**[2];
[2]Psychology, [1]Bowdoin Col., Brunswick, ME; [3]EpigenDx, Inc., Hopkinton, MA

Abstract: Early life adversity (ELA) significantly increases an individual's risk of developing affective disorders later in life, and interactions between genes and environment are key in facilitating - and mitigating - later life outcomes. Notably, women are at an increased risk of developing affective disorders, such as anxiety, following ELA compared to men. Treatment options for affective disorders are limited, as the complexity of underlying biochemical processes associated with how early life experience shape later life behavior is not well understood. The investigation of ELA-derived modifications to DNA provides an avenue through which these interactions can be explored to understand how epigenetics may serve to promote risk or resilience to adversity. We can leverage model systems of ELA - namely, the well characterized maternal separation (MS) model in rats - to identify links between alterations in the genome and concomitant aberrant behavior. In rats, ELA via MS leads to sex- and age-specific outcomes on brain and behavior comparable to those seen in human populations with a history of adversity. Here, we used the MS model to determine sex-specific developmental outcomes of ELA on epigenetic patterning in parvalbumin (PV) inhibitory neurons and behavioral output in a novel aversive ultrasonic vocalization (USV) playback assay of hypervigilance. We found that ELA leads to decreases in *Pvalb* mRNA, PV protein intensity, and PV cell count, with a parallel increase in intensity of DNA methylation (via 5-

methylcytosine (5-mC)) in prefrontal PV neurons; findings that were specific to juvenile female rats that had experienced ELA. Interestingly, we also observed a significant effect of ELA in the aversive USV playback task, where juvenile ELA females show a significant increase in anxiety-like behavior relative to other groups. Pyrosequencing and targeted bisulfite sequencing of several CpG sites on the *Pvalb* promotor also show sex-specific effects of ELA. Finally, we present preliminary evidence that 15mg/kg ketamine treatment reduces intensity of 5-mC signal in PV cells, which is specific to ELA females. Together, these findings suggest that PV-associated changes in DNA methylation may be involved in sex-specific anxiety-like behavior and could underly the sex-specificity of ELA-associated outcomes.

Disclosures: **E.S. Noel:** None. **Y. Peña:** None. **S.M. Bonauto:** None. **A. Chen:** None. **S. Ellis:** None. **A. Hopkins:** A. Employment/Salary (full or part-time); Employee of EpigenDx. **S. Miller:** A. Employment/Salary (full or part-time); Employee of EpigenDx. **J.A. Honeycutt:** C. Other Research Support (receipt of drugs, supplies, equipment or other in-kind support); Received assistance with analyses and sample processing from EpigenDx.

Poster

PSTR159. Early-Life Stress: Molecular Mechanisms and Cellular Effects

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR159.08/HH20

Topic: F.03. Stress and the Brain

Support: NIH Grant 1R01GM135247-01
NIH Grant R01 DA08259
Hope for Depression Research Foundation

Title: Single-cell genomic variation of functionally distinct hippocampal cell types in a mouse model of early life stress

Authors: *J. MARROCCO[1,2,3], A. JOGLEKAR[2], S. G. CARADONNA[3], J. JARROUX[2], R. WEINBERGER-GOLDBERG[2], S. MAZID[2], T. A. MILNER[2], H. U. TILGNER[2];
[1]Touro Univ., New York, NY; [2]Feil Family Brain and Mind Res. Inst., Weill Cornell Med., New York, NY; [3]The Rockefeller Univ., New York, NY

Abstract: Stress during the early postnatal period alters both the genome and function of the limbic system in humans and other species. The investigation of genomic signatures in brain circuits that regulate stress-related behaviors have been largely examined in tissues with cellular heterogeneity. We sought to investigate the genetic contribution of functionally distinct cell types within the ventral hippocampus (vHPC) in a mouse model of early life stress (ELS), which consists of limiting bedding and nesting material in the early postnatal period. After weaning, we found that ELS males, but not females, displayed lower body weight and shorter anogenital distance than controls. ELS also delayed the onset of vaginal opening. At postnatal day 29, brains were collected and coronal sections of the vHPC were either processed for single-cell

RNA sequencing or emersion fixed with 4% paraformaldehyde and processed for RNAscope in situ hybridization. Cell-by-gene expression matrices were generated to investigate the gene expression profile of each cell type. The Seurat R package for Model-based Analysis of Single Cell Transcriptomics (MAST) was used to find differentially expressed genes (DEGs) between experimental groups. We found sex-specific and ELS-dependent DEGs in functionally distinct cell types of the vHPC, namely excitatory pyramidal neurons, inhibitory neurons, granule neuroblasts, fibrous astrocytes, microglia, oligodendrocytes, and vascular endothelial cells. Several DEGs were involved in immediate signaling responses (*Egr1*) and excitatory neurotransmission (*Gria1*, *Grin2a*), which are major processes that coordinate neuroplasticity. In situ analyses showed downregulation of *Gria1* in pyramidal and granule cells, as well as in interneurons of ELS males and ELS females compared to controls. Contrariwise, *Grin2a* and *Egr1* were upregulated in pyramidal cells of ELS males compared to controls. In addition, the expression of *Grin2a* and *Egr1* was respectively upregulated in hilar interneurons and in granule cells of ELS males compared to controls. These findings demonstrate that the regulation of sex- and ELS-specific DEGs diverges in functionally distinct cell types of the vHPC despite genes clustering in similar biological processes. This indicates that the genome is programmed in early life at the gene-by-cell level, suggesting the need to integrate the genetic contribution of single cells to validate biomarkers of stress in genome-scale investigations.

Disclosures: **J. Marrocco:** None. **A. Joglekar:** None. **S.G. Caradonna:** None. **J. Jarroux:** None. **R. Weinberger-Goldberg:** None. **S. Mazid:** None. **T.A. Milner:** None. **H.U. Tilgner:** None.

Poster

PSTR159. Early-Life Stress: Molecular Mechanisms and Cellular Effects

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR159.09/HH21

Topic: F.03. Stress and the Brain

Support: NICHD 1R01HD087509-01
University of Delaware Winter Fellows Scholarship

Title: Consequences of early life stress on the epigenetic landscape of the developing brain: a role for exercise intervention

Authors: *A. L. SKWERES[1], T. CAMPBELL[2], K. DONOGHUE[2], T. ROTH[2];
[1]Psychological and Brain Sci., Univ. of Delaware Grad. Program In Behavioral Neurosci., Newark, DE; [2]Psychological and Brain Sci., Univ. of Delaware, Newark, DE

Abstract: Aversive caregiving in early life is a risk factor for aberrant brain and behavioral development throughout the lifespan.This outcome is related to epigenetic dysregulation of the *Bdnf* gene.The *Bdnf* gene encodes for brain-derived neurotrophic factor (BDNF),a neurotrophin involved in early brain development, neural plasticity, learning, and memory. Disruptions in

caregiver-infant interactions lead to increased methylation and decreased expression of the *Bdnf* gene. Aerobic exercise is hypothesized to mitigate this effect. Exercise increases BDNF at the protein and gene expression levels, making it an exciting target for therapeutic interventions. To our knowledge, exercise has never been studied as a therapeutic intervention in preclinical rodent models of early life caregiver maltreatment. To that end, the current study is investigating the effect of an adult voluntary wheel running intervention on *Bdnf* aberrant methylation and expression in the prefrontal cortex, cerebellum, and hippocampus of rats who experienced aversive caregiving in infancy. We employ a rodent model wherein rat pups experience intermittent caregiver-induced stress from postnatal days 1-7and are given voluntary access to a running wheel (except in the control condition)from postnatal days 70-89as a young adulthood treatment intervention. This study presents novel findings on the mechanisms underlying neural plasticity in brain regions critical for executive functioning, learning, and memory in an early life stress context. Current results suggest that exercise affects *Bdnf* gene methylation in an exon and CG site specific manner. Within ventral hippocampal tissue we report that *Bdnf* exon IV methylation is significantly lowered by wheel running compared to standard housed rats at the whole exon level [F(1,59)= 4.109,p= 0.0338].This effect is driven by methylation changes at the CG site-specific level, with wheel running significantly affecting methylation at CG sites 3,5, and 11([F(1,78)= 5.242,p=0.0248];[F(1,79)= 0.0296,p= 0.0296];[F(1,76)= 5.857,p= 0.018],respectively)of the exon IV promoter region. Sidak's multiple comparisons test revealed that wheel running lowered methylation at all three individual sites (p= 0.0472;p= 0.0198;p=0.017, respectively).Ongoing data collections and analyses are further probing this relationship to determine 1)how alterations in gene methylation may be related to changes in gene expression, 2)if a similar relationship between exercise and *Bdnf* epigenetic regulation is found in the cerebellum and prefrontal cortex, and 3)how exposure to early life stress may moderate the positive epigenetic effects of exercise.

Disclosures: **A.L. Skweres:** None. **T. Campbell:** None. **K. Donoghue:** None. **T. Roth:** None.

Poster

PSTR159. Early-Life Stress: Molecular Mechanisms and Cellular Effects

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR159.10/HH22

Topic: F.03. Stress and the Brain

Support: JSPS Grant 21K21124
JSPS Grant 23K16378
Grant from Dental Research Center, Nihon University School of Dentistry
Sato Fund, Nihon University School of Dentistry

Title: Increasing of copy number of LINE-1 in genomic DNA by maternal separation stress in mice

Authors: *M. OKANO, A. OGATA, M. KONDO;
Dept. of Legal Med., Nihon Univ. Sch. of Dent., Tokyo, Japan

Abstract: Although child maltreatment is a social issue, molecular biological markers have not been developed for its assessment. LINE-1 (long interspersed nuclear element-1: L1), which occupies more than 10% of genomic DNA, is known to insert into the genome due to its reverse transcription activity. In this study, we examined the association between stress exposure due to maternal separation of mice and the copy number of L1 in genomic DNA derived from blood. A total of eight pregnant mice were divided into two groups (low and high-stress groups). After parturition, the maternal separation was performed daily from the first day after birth (P1) to two weeks (P14). The low-stress group was exposed to maternal separation for 2 hours daily, and the infant mice were housed with others during treatment. On the other hand, high-stress group was conducted for 6 hours per day and the littermates were reared individually during maternal separation. Genomic DNA was extracted from the blood of euthanized infants after the treatment period (P15). Relative quantifications were then performed using 6 primer sets that were designed at three L1 subfamilies (Tf, A and Gf). In the results, the low-stress group showed no change against untreated mice (control) in littermate. In contrast, the high-stress group indicated copy number increases significantly in four of the six sites of L1. Specifically, the fold changes relative to control mice were 1.33 ($p < 0.001$) in one site of L1Gf, 1.11 ($p = 0.0099$) and 1.23 ($p = 0.0296$) in L1A and 1.40 ($p = 0.0051$) in L1Tf. The results suggested that the L1 copy number was increased according to the stress severity caused by maternal separation. Recently, schizophrenia has been observed as one of the long-term effects of child maltreatment whose severity is reported to be associated with the increasing copy number of L1 in genomic DNA (Bundo et al. 2014). Future work will be aimed at elucidating the relationship between L1 copy number and stress exposure due to maternal separation.

Disclosures: M. Okano: None. **A. Ogata:** None. **M. Kondo:** None.

Poster

PSTR159. Early-Life Stress: Molecular Mechanisms and Cellular Effects

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR159.11/HH23

Topic: F.03. Stress and the Brain

Support: NIH grant HD091376

Title: Stress during puberty produces lasting epigenetic changes in the brain that underlie risk for negative outcomes

Authors: *L. A. M. LUTHER, S. L. HIGLEY, K. N. GAUTIER, K. E. MORRISON;
West Virginia Univ., Morgantown, WV

Abstract: Experiencing adverse events during the critical developmental period of puberty puts women at risk for mood disorders in adulthood, especially if they become pregnant. We have

previously shown that undergoing stress during puberty and later becoming pregnant alters the response of the hypothalamic-pituitary-adrenal (HPA) axis, a key player in mood disorders, in adulthood in both humans and mice. We found that pubertal stress permanently altered gene expression in the brain region responsible for initiating the HPA axis, the paraventricular nucleus of the hypothalamus (PVN). We found an increased expression of six immediate early genes (IEGs) in the PVN of pregnant adult mice that had previously undergone pubertal stress. IEGs are stimulus-responsive genes that initiate downstream cellular cascades. However, these genes were permissively expressed in pregnant, pubertally stressed females in baseline conditions, suggesting a potential mechanistic role of IEGs in the blunted HPA axis phenotype. Further, ATAC-sequencing data showed that the lasting changes in gene expression are associated with alterations to the chromatin landscape, such that pregnant, pubertally-stressed females had increased openness of chromatin. Further analysis implicated histone acetylation in the increased openness. Here, we aimed to further understand these changes to the epigenome and the transcriptome. Beginning at postnatal day (PN) 21, mice were exposed to 14 days of chronic variable stress (CVS). Brains were collected either prior to stress (PN21), at the end of stress (PN35), or from adult, pubertally-stressed mice that were either pregnant or virgin. In late pregnancy, brains were collected from pregnant female and age-matched virgin females. H3K9 acetylation was quantified from PVN tissue. We found an association between pubertal stress exposure and H3K9 acetylation in adulthood. With the data from the PN21 and PN35 PVN, these results further our understanding of the specific epigenetic modifications caused by pubertal stress and their developmental trajectory. To better understand the role of the IEGs, we performed spatial transcriptomics for Fos mRNA in GABA and CRF reporter mice. These data provide a cell-type specific analysis of one of the key IEGs that we have found to be related to the blunted HPA axis. Altogether, these findings will clearly define the epigenetic plasticity that permits the blunted HPA axis response in pubertally-stressed females during pregnancy.These studies provide novel insight into the epigenetic mechanisms underlying female-relevant risk for stress dysregulation, a central endophenotype of affective disorders.

Disclosures: L.A.M. Luther: None. **S.L. Higley:** None. **K.N. Gautier:** None. **K.E. Morrison:** None.

Poster

PSTR159. Early-Life Stress: Molecular Mechanisms and Cellular Effects

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR159.12/HH24

Topic: F.03. Stress and the Brain

Title: Impact of an early life, social isolation mouse model on myelination, oligodendrocyte development, and cholesterol metabolism in the prefrontal cortex

Authors: *A. M. DAVISON, R. RAJU, J. F. SANTOYO, S. BARKER, N. E. MILMAN, L. A. AKAY, L.-H. TSAI;
Brain and Cognitive Sci., MIT, Cambridge, MA

Abstract: Impact of an early life, social isolation mouse model on myelination, oligodendrocyte development, and cholesterol metabolism in the prefrontal cortex

Authors* ***Alexis Davison***[1,2], *Ravikiran Raju*[1,2,3], *Juan Santoyo*[1,2], *Scarlett Barker*[1,2], *Noah Milman*[1,2], *Leyla Akay*[1,2], *Li-Huei Tsai*[1,2]

[1]*Picower Institute for Learning and Memory, Massachusetts Institute of Technology, Cambridge, MA, USA*[2]*Department of Brain and Cognitive Sciences, Massachusetts Institute of Technology, Cambridge, MA, USA*[3]*Division of Newborn Medicine, Boston Children's Hospital, Harvard Medical School, Boston, MA, USA*

DisclosuresNone.

Abstract It is a pivotal time to investigate the underlying neurobiological mechanisms impacted by social isolation. As the COVID-19 pandemic forced unnatural social isolation, the World Health Organization reported a 25% global surge in anxiety and depression cases. The prefrontal cortex (PFC) plays a vital role in responding to this stress. Previously, human studies showed less white cortical matter volume in orphans institutionalized at young ages (*Sheridan et al., 2012*), and mouse studies revealed less myelination in the PFC after an early critical period of isolation (*Makinodan et al., 2012*). We studied the impact of four weeks of social isolation immediately after weaning in wildtype C57BL/6J mice. Behavioral assessment indicated center aversion in open field testing and avoidance to light in light-dark testing. Electron microscopy uncovered axons with significantly higher g-ratios (hypomyelination) in the PFC. Immunohistochemistry exhibited a significant reduction in the number of mature $Olig2^{+}/CC1^{+}$oligodendrocytes and detected higher levels of BODIPY cholesterol and neutral lipid intensity in the PFC. RNA-sequencing of Olig2-positive cells (frontal cortex) showed upregulation of the cholesterol biosynthesis pathway. These insightful findings are motivating our current studies investigating various targets and interventions within the myelination, oligodendrocyte, and cholesterol pathways that could potentially reverse the behavioral and molecular phenotypes observed.

Disclosures: A.M. Davison: None. **R. Raju:** None. **J.F. Santoyo:** None. **S. Barker:** None. **N.E. Milman:** None. **L.A. Akay:** None. **L. Tsai:** None.

Poster

PSTR159. Early-Life Stress: Molecular Mechanisms and Cellular Effects

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR159.13/HH25

Topic: F.03. Stress and the Brain

Support: Basic Science Research Program (2020R1A6A3A03037828) of National Research Foundation (NRF) of Korea
Institutional Strategic Support Fund 3 scheme (ISSF3) to Translational Research Exchange @ Exeter (ISSF3-TREE-Choi, 2022) funded by Wellcome Trust
Mireille and Dennis Gillings Foundation Grant

Title: Adulthood transcriptomic and behavioral alterations induced by developmental Glucocorticoid exposure in a zebrafish model

Authors: *M.-K. CHOI[1], A. COOK[2], H. EACHUS[1], A. TOCHWIN[1], S. RYU[1];
[1]Univ. of Exeter, Exeter, United Kingdom; [2]Inst. for human Genet., Univ. of Mainz, Mainz, Germany

Abstract: Chronic or severe stress during development has been linked to an increased risk of adult diseases in humans. One proposed mechanism involves the priming of stress-sensitive gene networks through early life exposure to glucocorticoids (GCs), which can modify stress responses later in life. However, the comprehensive understanding of brain-wide molecular alterations induced by GC priming remains limited. In this study, we utilized an optogenetic zebrafish model to elevate endogenous GC levels during development and characterized the transcriptomic changes in the brain across the life course. When developmental GC-exposed fish were exposed to acute stress in adulthood, they exhibited highly exaggerated endocrinal and transcriptional changes. These changes included gene sets associated with axon development, neuronal signaling, and epigenetic modulators. Remarkably, the altered gene sets were enriched with risk genes associated with human psychiatric disorders. Notably, subsets of primed genes and putative transcriptional regulators were found to be associated with changes in adult social behavior, including oxytocin and *myt1la*. Thus, our findings establish a novel and translationally relevant zebrafish model, providing valuable insights into the molecular targets through which developmental GC exposure may exert life-long impacts.

Disclosures: M. Choi: None. **A. Cook:** None. **H. Eachus:** None. **A. Tochwin:** None. **S. Ryu:** E. Ownership Interest (stock, stock options, royalty, receipt of intellectual property rights/patent holder, excluding diversified mutual funds); European patent number 2928288 and US patent number 10,080,355: “A novel inducible model of stress.”.

Poster

PSTR159. Early-Life Stress: Molecular Mechanisms and Cellular Effects

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR159.14/HH26

Topic: F.03. Stress and the Brain

Title: Effect of glucocorticoids on perineuronal nets expression in cultured cortical neurons

Authors: *L. YUE, B. MORRIS;
Univ. of Glasgow, Glasgow, United Kingdom

Abstract: Background: Schizophrenia is a complex psychiatric disorder which could be affected by environmental risk factors. Multiple studies have suggested that early developmental exposure to stressful events could increase the risk of schizophrenia. perineuronal nets (PNNs) are extracellular matrix structures surrounding mainly parvalbumin (Pv)-expressing GABAergic interneurons, providing a neuronal protective function during the developmental period. Recent

studies revealed that stress could alter the density and intensity of PNNs and disruption of PNNs could be observed in schizophrenia. However, the effect of stress on PNNs' specific components remained to be discovered. As glucocorticoids secretion could mediate the effect of stress, the current study aimed to investigate the effect of glucocorticoids on the expression of PNNs' components, including chondroitin sulfate proteoglycans (CSPGs), hyaluronan synthesis (Has), hyaluronan link proteins (Hapln) and tenascinR (TnR). **Methods**: primary mouse cortical cultures were treated with hydrocortisone acetate (HCA), mifepristone, collagenIII, aldosterone for 4h or 24h at 7, 14 and 21 days in vitro (DIV). The mRNA expression of PNNs components was examined using quantitative polymerase chain reaction (qPCR). **Results**: we found that mRNA expression of Pv, TnR and neurocan (Ncan) decreased at 7DIV, and the expression of Has1 and Has2 increased at 14DIV after cotreatment of HCA and mifepristone, indicating glucocorticoids could alter PNNs components through glucocorticoids receptors (GR). Additionally, we then treated the cells with collagenIII, a ligand of glucocorticoid-bound adhesion G-protein receptor (GPR) to test whether glucocorticoid could alter the PNNs expression via GPR except from GR. The results revealed that the expression of Ncan was regulated by collagenIII at 7DIV, suggesting that stress could alter Ncan's expression through GPR. Moreover, aldosterone, an agonist of mineralocorticoids receptors, was treated to the cultured cells to test whether glucocorticoids could affect PNNs' expression via MR. However, we found that the mRNA expression of Bcan, Vcan and Hapln4 remained unchanged after aldosterone treatment at 14DIV. **Conclusions**: the results suggested that high-dose glucocorticoids could alter the expression of TnR, Ncan, Pv and Has1 via glucocorticoids receptors and could also regulate the expression of Ncan via ADGRG3/GPR97. To conclude, the results suggested glucocorticoids might mediate early developmental stress effects on the expression of PNNs expression. Future studies aim to investigate the effect of glucocorticoids on the PNNs expression in the adult brain.

Disclosures: L. Yue: None. **B. Morris:** None.

Poster

PSTR159. Early-Life Stress: Molecular Mechanisms and Cellular Effects

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR159.15/HH27

Topic: F.03. Stress and the Brain

Support: DK124727
GM060507
MD006988

Title: Targeting Synaptic Vesicle 2A (SV2A) to Reduce Stress-Induced Binge Eating

Authors: *T. SIMON[1], V. WILLIAMS[1], A. WILLIAMS[1], J. D. FIGUEROA[2];
[2]Loma Linda Univ. Hlth., [1]Loma Linda Univ., Loma Linda, CA

Abstract: Background: Emotional eating and binge eating are coping strategies in response to social stressors early in life, resulting in overeating, enhanced intake of "comfort" foods high in fats and sugars, and obesity. However, the neurobiological mechanisms that link stress to maladaptive coping and disordered eating remain to be clarified. Evidence demonstrates that social isolation stress profoundly alters synaptic function and neuroplasticity. The Synaptic Vesicle 2A (SV2A) modulates neurotransmitter release in the presynaptic terminal of excitatory (E) and inhibitory (I) neurons, thus regulating the E/I balance in critical brain regions coordinating emotionality and eating. Therefore, we **hypothesize that restoring SV2A levels and synaptic integrity will confer resilience to social isolation stress and reduce binge-eating behaviors. Methods:** Adolescent Lewis rats (Male = 32, Female = 32) were weaned at postnatal day (PND) 21 and randomly allocated to one of two groups based on housing conditions: Paired or Isolated. The Paired animals were housed in two rats per cage, while the Isolated animals were single-housed throughout the study. After adolescence (PND 61), all animals underwent a battery of behavioral tests to assess startle reactivity, anxiety-like behaviors, and sociability. After the behavioral tests, all the rats underwent a binge eating paradigm consisting of three weekly short-term exposures to a Western-like diet (*WD*, 41% kcal from fat). In the third week, the Paired and Isolated groups were subdivided into Vehicle and the anticonvulsant drug, Levetiracetam (LEV) (n = 8). LEV (10 mg/kg via intraperitoneal injection) was administered before the third WD exposure, and food consumption was monitored. Brain tissue was harvested at PND 105 and prepared for the immunohistological evaluation of SV2A and cellular phenotypes. **Results:** Results suggest that adolescent social isolation stress leads to elevated weight gain/food consumption, stress reactivity, and anxiety while reducing sociability in adult rats. Notably, LEV
administration modulated feeding behaviors and reduced binge-like eating in female rats. This study may aid in developing clinically relevant treatments for stress-related eating disorders.

Disclosures: T. Simon: None. **V. Williams:** None. **A. Williams:** None. **J.D. Figueroa:** None.

Poster

PSTR159. Early-Life Stress: Molecular Mechanisms and Cellular Effects

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR159.16/HH28

Topic: F.03. Stress and the Brain

Support: NSF IOS-1929829
NIH MH129020
NIH DA049837
NIH DA056534
NIH DA055846
NSF 1625061

Title: Early life adversity increases cortical astrocyte volume and impacts the transcriptome of the orbitofrontal cortex in adult male and female rats

Authors: *E. HARRIS[1], C. DECKERS[2], R. KARBALAEI[2], N. MILES[2], E. HARDER[3], E. WITT[3], K. J. REISSNER[3], M. E. WIMMER[2], D. A. BANGASSER[1];
[1]Ctr. for Behavioral Neurosci., Georgia State Univ., Atlanta, GA; [2]Dept. of Psychology and Neurosci. Program, Temple Univ., Philadelphia, PA; [3]Dept. of Psychology and Neurosci., UNC Chapel Hill, Chapel Hill, NC

Abstract: Astrocytes are glial cells in the brain important for homeostatic functions, regulating synaptic transmission, synapse formation and remodeling, and metabolism. Astrocytic morphology is particularly key to their function and ability to properly modulate synaptic activity. Previous studies have demonstrated that astrocytic morphology is particularly impacted by acute and chronic stress paradigms in adulthood. Here we sought to understand whether early adversity can impact the morphology of astrocytes within key brain regions. Our laboratory uses the limited bedding and nesting (LBN) model, where dams and pups experience a low resource environment from pups' postnatal day 2-9. LBN fragments maternal care and increases dams' pup-directed behavior at the expense of their self-care. Previously we reported that LBN affects cognitive tasks that are primarily controlled by the medial prefrontal cortex (mPFC) and orbitofrontal cortex (OFC), two areas where adult stressors can affect astrocyte morphology. First, we used a novel viral approach that fully labels astrocyte morphology and found that LBN increases the volume and surface area of astrocytes in mOFC and mPFC in adult male and female rats. These morphological changes were similar across sex. Next, we performed RNA sequencing on OFC tissue to uncover potential mechanisms by which LBN can alter astrocytes. We found there was sex-specific regulation of the OFC transcriptome by LBN. One pathway altered by LBN in both sexes was glutamate signaling, although the changes in transcripts in that pathway were sex-specific. Glutamate alters astrocyte morphology, so it is possible that sex-convergent changes in glutamatergic signaling due to LBN account for the increased astrocyte morphology. Furthermore, we identified specific sub-cell types that may be responsible for driving the changes observed in a bulk RNA-seq differential gene expression experiment using the LRcell deconvolution package**.** This analysis identified astrocytes as one of the sub-cell types significantly affected by LBN in the OFC of males and females. Given the role of astrocytes in a range of functions, changes in their morphology and gene expression profile likely have a significant impact on cortical brain function and behavior.

Disclosures: E. Harris: None. **C. Deckers:** None. **R. Karbalaei:** None. **N. Miles:** None. **E. Harder:** None. **E. Witt:** None. **K.J. Reissner:** None. **M.E. Wimmer:** None. **D.A. Bangasser:** None.

Poster

PSTR159. Early-Life Stress: Molecular Mechanisms and Cellular Effects

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR159.17/II1

Topic: F.03. Stress and the Brain

Support: CIHR grant PJT-173287

Title: Investigating perineuronal nets and myelination around parvalbumin interneurons in human ventromedial prefrontal cortex

Authors: *S. THÉBERGE, C. BELLIVEAU, D. XIE, R. KHALAF, M. DAVOLI, N. MECHAWAR;
Douglas Mental Hlth. Univ. Inst., Verdun, QC, Canada

Abstract: Background During development, critical periods (CP) are important windows of plasticity during which cortical circuits are shaped more easily by the environment. Perineuronal nets (PNN) are key regulators of neuroplasticity and their postnatal maturation around parvalbumin (PV) interneurons is associated with closing this period of heightened plasticity. Myelination also plays a role in halting CPs, and recent studies have revealed that cortical PV axons are myelinated. Previous findings from our lab have shown that a history of severe child abuse (CA) is associated with impaired myelination in the anterior cingulate cortex (ACC) and increased recruitment and maturation of PNNs in the ventromedial prefrontal cortex (vmPFC). This project tests the hypotheses that there is a relationship between PNNs and myelination around PV interneurons, and that severe CA affects this relationship. **Methods** Well-characterized post-mortem human vmPFC samples from adult male and female depressed suicides with or without a history of severe CA (DS-CA & DS) and matched healthy controls (CTRL) were acquired from the Douglas-Bell Canada Brain Bank (n=36). These samples were matched for age, post-mortem interval, and brain pH. We used immunofluorescent labeling with three primary antibodies directed against PV, Myelin Basic Protein (MBP), and Neurofascin (axon initial segments), as well as with Wisteria Floribunda Lectin to stain PNNs. Multiarea timelapse z-stack image acquisitions were performed on a confocal microscope with a 60x objective. Analysis of the relation between PNN coverage and PV axon myelination was performed using the plugin Simple Neurite Tracer (SNT) from FIJI. All analyses regarding the nature of the samples (CTRL, DS, and DS-CA) were done in a blinded fashion. **Results** We found that 81% of PV axons were myelinated and that 55% were covered by PNNs in CTRLs. In CTRLs, most PV cells (52%) are myelinated with a PNN, with few being unmyelinated with a PNN (3%). In DS-CA, preliminary results reveal a decreasing trend in the proportion of PV interneurons with a myelinated axon, along with an increase in the proportion of unmyelinated PV interneurons covered by a PNN compared to DS and CTRL. **Conclusion** Together, these preliminary results suggest a potential relationship between PNNs and PV interneuron myelination and should shed new light on possible CA-associated changes in myelination and PNN formation.

Disclosures: S. Théberge: None. **C. Belliveau:** None. **D. Xie:** None. **R. Khalaf:** None. **M. Davoli:** None. **N. Mechawar:** None.

Poster

PSTR159. Early-Life Stress: Molecular Mechanisms and Cellular Effects

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR159.18/II2

Topic: F.03. Stress and the Brain

Support: CIHR grant PJT-156346

Title: Examining Neurovascular Dysfunction in Depressed Suicides with a History of Childhood Abuse

Authors: *M. WAKID[1,2], D. ALMEIDA[1,2], Z. AOUABED[1], R. DENNISTON[1], R. RAHIMIAN[1], V. YERKO[1], E. LEONOVA-ERKO[1], M. DAVOLI[1], G. TURECKI[1], N. MECHAWAR[1];
[1]Douglas Mental Hlth. Univ. Inst., Montreal, QC, Canada; [2]McGill Univ., Montreal, QC, Canada

Abstract: Introduction: Childhood abuse (CA), is experienced globally by approximately 1 billion youth aged between 2-17 years (Hillis et al., 2016, Pediatrics, 137:1). Consistent evidence of neuroinflammation and activated microglia (Lehmann et al., 2018, Scientific Reports, 8:5) but also microbleeds (Lehmann et al., 2018, Scientific Reports, 8:5), abnormal blood vessel morphology and downregulated tight junction protein claudin5 (Cldn5) are observed in the brains of mice exposed to chronic stress (Menard et al., 2017, Nat Neurosci, 20:3; Dudek et al., 2020, Proc Natl Acad Sci USA, 117:2). However, the impact CA has on cerebrovascular integrity has yet to be thoroughly investigated in humans. Hypothesis & Objectives: We hypothesize that a history of early-life stress exacerbates alterations in neurovasculature, as suggested by previous studies of chronic stress experienced in adulthood, with significant changes experienced in the ventromedial prefrontal cortex (vmPFC). Methods: Well-characterized frozen postmortem brain samples from adult male and female depressed suicides with a history of severe CA and matched sudden-death controls (n=26/group) were obtained from the Douglas-Bell Canada Brain Bank. We developed a protocol to enrich and isolate microvessels using mechanical homogenization and centrifugation-separation that is gentle enough to maintain the structural integrity and multicellular composition of intact microvessels from both groups for downstream investigation using RNA-sequencing. Results: Differential gene expression analysis revealed that, when compared to CTRLs, samples from CA victims exhibit dysregulation in several molecular functions at the neurovascular unit, including immune signaling (CD74, $p = 0.005$; CSF1, $p = 0.03$), glucocorticoid signaling (DNMT1, $p = 0.02$; HSP90B1, $p = 0.002$), and vascular biochemical activity (PTGS2, $p = 0.008$), indicating a role for neurovascular dysfunction in the pathological effects of chronic stress. Validation experiments (RNAscope and qPCR) are currently being conducted. Significance: To our knowledge, this is the first report of microvessels being effectively isolated from human brain tissue and analyzed as an intact structure to explore the latent neurovascular associated with a history of CA. Our results not only characterize the contributions of neurovascular dysfunction but highlight potential therapeutic targets at the blood-brain barrier.

Disclosures: M. Wakid: None. **D. Almeida:** None. **Z. Aouabed:** None. **R. Denniston:** None. **R. Rahimian:** None. **V. Yerko:** None. **E. Leonova-Erko:** None. **M. Davoli:** None. **G. Turecki:** None. **N. Mechawar:** None.

Poster

PSTR159. Early-Life Stress: Molecular Mechanisms and Cellular Effects

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR159.19/II3

Topic: F.03. Stress and the Brain

Support: CIHR grant PJT-156346

Title: An examination of ITGB1 expression in the hippocampus of depressed suicides with a history of child abuse

Authors: *R. CHEN[1,2,3], S. SIMARD[2,3], M. DAVOLI[2], N. MECHAWAR[2,3,4];
[1]Douglas Mental Hlth. Univ., Verdun, QC, Canada; [2]Douglas Mental Hlth. Univ. Inst., Verdun, QC, Canada; [3]Integrated Program in Neuroscience, McGill Univ., Montréal, QC, Canada; [4]Psychiatry, McGill Univ., Montréal, QC, Canada

Abstract: An examination of ITGB1 expression in the hippocampus of depressed suicides with a history of child abuse

Authors

***R. Chen**, S. Simard, M.A. Davoli, N. Mechawar

Disclosures

R. Chen: None. **S. Simard**: None. **M.A. Davoli**: None. **N. Mechawar**: None.

Abstract

Early life adversity (ELA) is the single largest risk factor for psychiatric disorders, yet much is still unknown about its lasting impacts on the brain. In both human and animal models, ELA has been shown to lastingly affect cortical oligodendrocyte-lineage cells and white matter. However, the regional specificity and molecular mechanisms underlying these effects remain to be explored. Our group previously reported a significant decrease in Integrin Beta Subunit 1 (ITGB1)expression in the anterior cingulate cortex of depressed suicides with a history of child abuse compared to matched controls. ITGB1 plays a pivotal role in the maturation and proliferation of oligodendrocytes. The current study is aimed at examining post-mortem ITGB1 expression in the hippocampus of individuals with a history of child abuse (CA; samples from the Douglas-Bell Canada Brain Bank). We first used spatial transcriptomics (Visium) to visualize the spatial expression of ITGB1 RNA and various oligodendrocyte markers at the subspot level in hippocampus samples from nonpsychiatric controls. ITGB1 was predominantly located in hippocampal white matter, overlapping in many subspot regions with early oligodendrocyte lineage markers. To investigate potential co-expression on a cellular level, we then used a quantitative RNAscope approach. Our preliminary data suggest high and widespread expression of ITGB1 throughout the hippocampus, particularly in the white matter, and co-expression of ITGB1 and PDGFRa was also mostly observed in white matter. Furthermore, preliminary cell counts indicated fewer ITGB1 and PDGFRa-positive OPCs expressing ITGB1 in depressed suicides with a history of CA compared to matched controls. Although there was

only a minor decrease in overall ITGB1 expression associated with CA, this observation can be explained by an increase in ITGB1 expression in non-OPC cells. These results suggest that the dysregulation caused by ITGB1 may depend not only on overall expression but also on the specific downregulation of ITGB1 inOPCs and other early oligodendrocyte lineage cells. We will also present data on hippocampal myelination, which we hypothesize, given disruptions in ITGB1, is altered in samples from individuals with a history of CA. Overall, these preliminary results suggest that ITGB1 is affected by childhood abuse in multiple brain regions and may impact cell types differentially.

Disclosures: **R. Chen:** None. **S. Simard:** None. **M. Davoli:** None. **N. Mechawar:** None.

Poster

PSTR159. Early-Life Stress: Molecular Mechanisms and Cellular Effects

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR159.20/II4

Topic: F.03. Stress and the Brain

Support: Canadian Institutes of Health Research (CIHR) grant PJT-156346
Canadian Institutes of Health Research (CIHR) Doctoral Award

Title: Fatty acid composition in phospholipid fractions of the uncinate fasciculus in depressed suicides

Authors: *K. PERLMAN[1], C. T. CHEN[2], M. SMITH[2], M. ORRI[1], G. TURECKI[1], R. P. BAZINET[2], N. MECHAWAR[1];
[1]Douglas Mental Hlth. Univ. Inst., Verdun, QC, Canada; [2]Univ. of Toronto, Toronto, ON, Canada

Abstract: Background: Major depressive disorder (MDD) is the leading cause of disability worldwide, with over 300 million people affected globally. White matter disruptions have been reported in MDD in both neuroimaging and molecular studies. Since the myelin sheath is highly enriched in lipids, white matter dysfunction may reflect alterations in the myelin lipid profile, given that the composition of fatty acids (FA) in myelin phospholipids influence its compactness, stability, and permeability. Numerous lipid-related abnormalities associated with MDD have been reported in the periphery, though very little information is available for the brain, especially for long-range white matter tracts. Therefore, the objective of this study is to quantify FA concentrations in the postmortem human uncinate fasciculus (UF), a major association tract, and characterize their relationships with MDD, child abuse (CA) history, and age. FA concentrations in all major phospholipid pools were compared between depressed suicides with a history of CA, depressed suicides without CA, and non-psychiatric controls. **Methods**: Group-matched brain samples were provided by the Douglas-Bell Canada Brain Bank. Total lipids were extracted according to the Folch method, lipids were separated into respective classes using thin-layer chromatography, and FA methyl esters from each fraction were quantified using gas

chromatography. **Results:** Phospholipid fractions revealed divergent patterns of FA composition (both in concentration and relative percentage) in MDD and CA samples compared to nonpsychiatric controls. In the sphingomyelin fraction only, the monounsaturated acid relative percentage is decreased in MDD compared to controls ($p = 0.0053$). Across phospholipid classes, the age relationships varied quite significantly, though there was some overlap, for example with arachidonic acid and other polyunsaturates. **Conclusion:** We present the first ever characterization of UF phospholipid FAs and describe their relationships with MDD, CA, and age. These data will be supplemented with cholesterol quantification and single nucleus gene expression data, to better understand their biological relevance.

Disclosures: K. Perlman: E. Ownership Interest (stock, stock options, royalty, receipt of intellectual property rights/patent holder, excluding diversified mutual funds); Aifred Health. **C.T. Chen:** None. **M. Smith:** None. **M. Orri:** None. **G. Turecki:** None. **R.P. Bazinet:** B. Contracted Research/Research Grant (principal investigator for a drug study, collaborator or consultant and pending and current grants). If you are a PI for a drug study, report that research relationship even if those funds come to an institution.; Arctic Nutrition, Bunge Ltd., DSM, Fonterra Inc, Mead Johnson, Natures Crops International, Nestec Inc. Pharmavite, and Sancero Inc.. **N. Mechawar:** None.

Poster

PSTR159. Early-Life Stress: Molecular Mechanisms and Cellular Effects

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR159.21/II5

Topic: F.03. Stress and the Brain

Support: CIHR Grant PJT-173287

Title: A cross-species characterization of perineuronal nets in the cerebellum

Authors: *R. MPAI[1], C. HERCHER[1], C. BELLIVEAU[2], M.-A. DAVOLI[3], N. MECHAWAR[4]; [1]McGill Univ. Integrated Program in Neurosci., Montreal, QC, Canada; [2]Douglas Hosp. Res. Ctr., Douglas Hosp. Res. Ctr., Montreal, QC, Canada; [3]Douglas Mental Hlth. Univ. Inst., Montréal, QC, Canada; [4]Douglas Inst., Douglas Inst., Montreal, QC, Canada

Abstract: Introduction: Perineuronal nets (PNNs) are structures of condensed extracellular matrix that have been shown to restrict neuroplasticity and stabilize synapses. Though they have been well-characterized in sensory cortices, little is known about PNNs in the cerebellum (CB), particularly in humans. The human cerebellum is believed to have expanded in parallel with the neocortex, possibly giving rise to uniquely human traits. Despite this expansion, the general organization of the cerebellum has remained conserved across species. This study aims to characterize CB PNNs through a cross-species comparison of mice, macaques and humans. Methods: Post-mortem human CB from neurologically and psychiatrically healthy individuals were provided by the Douglas-Bell Canada Brain Bank. CB from adult wild-type mice and

cynomolgus macaques were obtained through collaborations. Using immunofluorescence, we labelled PNNs using Wisteria Floribunda Lectin (WFL) and an antibody against aggrecan (ACAN). In the animal samples, we labelled parvalbumin(PV)-expressing neurons with an anti-PV antibody. Fluorescent in situ hybridization (FISH) was employed to label vesicular glutamate transporter 1 (SLC17A7), glutamate decarboxylase 1 (GAD1), and PV to determine the phenotype of cells surrounded by PNNs in the human deep CB nuclei (DCN). Preliminary results: We observed WFL+ and ACAN+ PNNs in the DCN across the species examined. In mice and macaques, we also found WFL+ and ACAN+ PNNs in the CB cortex with differences in expression between markers. In the mouse CB, no PNNs were surrounding PV+ neurons. In macaques, PNNs in the CB nuclei mostly surrounded PV+ neurons, while those in the CB cortex were mostly PV-. FISH experiments revealed that human CB PNNs mostly surround PV+/SLC17A7+ neurons. Conclusion: This study will add to the growing body of literature on the topic of PNN expression in post-mortem human tissue**.** Moreover, this work highlights species differences in the nature and distribution of CB PNNs and paves the way for future studies on PNN-related CB neuroplasticity in the healthy and disordered brain.

Disclosures: R. Mpai: None. **C. hercher:** None. **C. Belliveau:** None. **M. Davoli:** None. **N. Mechawar:** None.

Poster

PSTR159. Early-Life Stress: Molecular Mechanisms and Cellular Effects

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR159.22/II6

Topic: F.03. Stress and the Brain

Support: The Ohio State University Chronic Brain Injury Institute seed grant
Brain Injury Association of America Seed Grant
The Ohio State University Chronic Brain Injury Institute pilot funding
NSF Graduate Research Fellowship DGE-1343012
The Ohio State University Chronic Brain Injury Institute Summer Undergraduate Research Fellowship

Title: Examining the impact of early life stress and pediatric TBI on the developing hippocampal transcriptome and behavioral development in rats.

Authors: *M. R. BREACH[1], E. GOODMAN[2], J. PACKER[2], A. ZALETA LASTRA[1], H. E. AKOURI[1], Z. M. TAPP-POOLE[2], C. VONDER HAAR[2], J. GODBOUT[2], O. KOKIKO-COCHRAN[2], K. M. LENZ[1];
[1]Psychology, [2]Neurosci., The Ohio State Univ., Columbus, OH

Abstract: Background: Adverse childhood experiences (ACEs) increase one's lifetime risk for developing neuropsychiatric disorders. ACEs include psychosocial stress and physical violence, which may result in pediatric traumatic brain injury (TBI). Pediatric TBI independently increases

one's risk for impaired neuropsychiatric functioning. Despite the prevalence and substantial cooccurrence rates of ACEs and pediatric TBI, few rodent studies have assessed how chronic stress modifies the impact of pediatric TBI. Here, we developed a 'two hit' rat model to explore whether stress experience modifies the impact of TBI on the brain and behavior. **Methods:** From postnatal day (P)1-14, rats underwent daily handling or maternal separation stress for 4hr. Then, rats were subjected to either a lateral fluid percussion injury or isoflurane anesthesia on P15. At 7 days post injury (DPI), ipsilateral hippocampus was dissected from a subset of males (n = 3/group). Pooled samples were processed for nuclei isolation, fixation, and barcoding. Single nucleus RNA sequencing was conducted on ~30,000 nuclei and Ingenuity Pathway Analyses were performed on significant differentially expressed genes. Remaining rats (n = 6-11/group, both sexes) were assessed for open field behavior, spontaneous alternation, and spatial reference memory in adulthood (≥ 53 DPI) by an experimenter blind to condition. **Results:** Stress and Stress + TBI activated pathways associated with plasticity in excitatory and inhibitory neurons, including long-term potentiation and long-term depression. Stress and Stress + TBI treatment also activated neuromodulator signaling pathways, including those of oxytocin, opioids, and dopamine. TBI activated plasticity-associated pathways in excitatory neurons only. In microglia, stress generally deactivated phagocytic and inflammatory pathways, and this was exacerbated by combination with TBI. Upstream regulator analysis found that *CSF3*, *GRIN3A*, and *MECP2* may regulate gene expression changes across adversity models. However, *DAG1* was uniquely implicated in neuronal populations for the Stress + TBI group. Regarding behavioral analyses, stress increased exploratory risk-taking behavior in the open field. **Conclusion:** Early life stress profoundly impacts the juvenile hippocampal transcriptome and later life exploratory behavior. Combination with TBI alters hippocampal transcriptomic effects. Future work will add females to the sequencing analysis to determine if this model modulates gene expression in a sex-specific manner. We will apply the knowledge obtained from these analyses to assess mediators of adversity-induced neurobehavioral dysfunction.

Disclosures: **M.R. Breach:** None. **E. Goodman:** None. **J. Packer:** None. **A. Zaleta Lastra:** None. **H.E. Akouri:** None. **Z.M. Tapp-Poole:** None. **C. Vonder Haar:** None. **J. Godbout:** None. **O. Kokiko-Cochran:** None. **K.M. Lenz:** None.

Poster

PSTR159. Early-Life Stress: Molecular Mechanisms and Cellular Effects

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR159.23/II7

Topic: F.03. Stress and the Brain

Support: CIHR grant PJT-173287

Title: Child abuse-associated changes in prefrontal cortical perineuronal nets: investigating the implication of microglia

Authors: *C. BELLIVEAU, R. RAHIMIAN, A. TANTI, S. THÉBERGE, C. HOSDEY, R. MPAI, G. FAKHFOURI, M. DAVOLI, N. MECHAWAR;
Douglas Hosp. Res. Ctr., Montreal, QC, Canada

Abstract: Background: Severe abuse experienced during the critical period of developmental brain plasticity significantly increases the risk of depression and suicide later in life. In this study, we investigated the impact of child abuse (CA) on the interaction between microglia (MG) and perineuronal nets (PNNs) in the adult ventromedial prefrontal cortex (vmPFC). Our previous research revealed an association between CA and enhanced recruitment and maturation of PNNs in depressed suicides compared to matched controls. While recent literature suggests that MG are involved in the maintenance and degradation of PNNs, the underlying mechanisms remain unclear. **Methods**: To explore this further, we obtained well-characterized human vmPFC samples (N=37) from the Douglas-Bell Canada Brain Bank along three groups comprising both males and females: depressed suicides with (DS-CA) and without (DS) a history of child abuse to psychiatrically healthy controls. We also investigated mPFC samples from adult male and female C57BL/6 mice (N=19) subjected to limited bedding and nesting during early postnatal development (PD2-9). Immunofluorescence and in situ hybridization were employed to examine the direct spatial relationship between MG and PNNs in both human and mouse samples. Matrix metalloproteinase (MMPs) antibody arrays, ELISA, and immunoblotting were used to explore the indirect regulation of PNNs by MG through enzyme secretion. **Results**: Our spatial analysis suggests that the direct regulation of PNNs by MG does not differ between groups in both human and mouse samples. However, MG appear to play an indirect role in maintaining the increased recruitment of PNNs in the vmPFC of DS-CA, as demonstrated by a significant downregulation of MMP-9 in bulk grey matter and MG (CD11b+ pulldown). Additionally, significantly decreased levels of Cathepsin S, CX3CR1, and the neo-epitope of aggrecan cleaved by MMPs were observed in DS-CA samples, further highlighting a dysregulation in the indirect MG-PNN relationship. **Conclusion**: This study provides the first evidence in humans that MG have an indirect involvement in the maintenance of increased PNNs in the vmPFC of individuals with a history of CA. Furthermore, our findings are supported by a mouse model of early-life adversity. Understanding the long-lasting impacts of CA on the brain is crucial for reducing the global burden of depression and suicide.

Disclosures: C. Belliveau: None. **R. Rahimian:** None. **A. Tanti:** None. **S. Théberge:** None. **C. Hosdey:** None. **R. Mpai:** None. **G. Fakhfouri:** None. **M. Davoli:** None. **N. Mechawar:** None.

Poster

PSTR159. Early-Life Stress: Molecular Mechanisms and Cellular Effects

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR159.24/II8

Topic: F.03. Stress and the Brain

Support: RGPIN-2018-05063

Title: Medial prefrontal cortex transcriptomics unveils molecular mechanisms linked to the sex-specific effects of prenatal adversity on risk-taking behavior

Authors: *P. M. MIGUEL[1,3], B. BARTH[2], M. B. ALVES[1], R. DALLE MOLLE[4], A. GÓMEZ-ILESCAS[2], A. BATRA[2], D. M. ARCEGO[5], T. ZHANG[5], X. WEN[4], C. PARENT[5], N. O'TOOLE[5], M. J. MEANEY[1,6], P. P. SILVEIRA[1,7,8];
[1]Dept. of Psychiatry, [2]Integrated Program in Neurosci. (IPN), McGill Univ., Montreal, QC, Canada; [3]Dept. of Psychiatry, McGill Univ., Montreal, QC, Canada; [5]Ludmer Ctr. for Neuroinformatics and Mental Hlth., [4]Douglas Res. Ctr., Montreal, QC, Canada; [6]Agency for Science, Technol. and Res. (A*STAR), Singapore Inst. for Clin. Sci., Singapore, Singapore; [7]Integrated Program in Neurosci. (IPN), McGill Univ., Montreal, QC, Canada; [8]Ludmer Ctr. for Neuroinformatics and Mental Health, Douglas Res. Ctr., Montreal, QC, Canada

Abstract: Prenatal adversity modifies individual responses to environmental cues, which can lead to risk-taking behavior and the development of chronic diseases over the life course. We have described, in an animal model, that prenatal adversity changes offspring dopamine release in the medial prefrontal cortex (mPFC), inducing increased impulsive behavior in females. Here, we investigated risk-taking behavior and biological pathways associated with these effects by RNA-sequencing analyses in the mPFC of males and females rats exposed to prenatal adversity. On pregnancy day 10, dams were allocated to control (*ad libitum* diet) or food restriction (FR) group (50% restricted diet). At birth, all pups were adopted by control dams aiming at isolating the effect of FR prenatally. At postnatal day 90 (P90), the offspring risk-taking behavior was measured in two cohorts using distinct behavioral tasks with aversive conditions to open and bright areas: the light-dark box (LDB; n=7-8/group) and the novelty-suppressed feeding (NSF; n=17-20/group). Another cohort of FR animals was euthanized on P90, and the mPFC was collected to perform RNA-sequencing to identify differentially expressed genes (DEGs) in males and females separately. In the LDB, an increase in time exploring the aversive compartment, and in the number of crossings between light/dark compartments were observed in FR animals, independent of sex. Additionally, FR animals started to eat faster the palatable food in the center of the aversive arena in the NSF, with no effects on the latency to eat in their home cage. Interestingly, FR females ate larger amounts of palatable food in the arena and lost more body weight due to fasting before the test. Group and sex effects were also observed in total distance traveled and average speed, indicating that FR and female rats increased their locomotor activity in the arena. FR induced 759 DEGs in the mPFC of females and 670 in males, with only 46 in common between both sexes. DEGs present only in the mPFC of FR females are enriched for more unique pathway maps (such as the Notch signaling), for processes associated with neuronal development and cellular organization, and for neurodevelopmental disorders and neurological manifestations, when compared to DEGs from males. Our results demonstrated that prenatal adversity induced sex-specific DEGs in the mPFC, which mapped into specific molecular pathways, that can be associated with the pronounced effect in risk-taking behavior in females. Since scanning the environment in a proper way is important for adaptation and survival, our findings indicate a disruption in this behavior that could be linked to the development of chronic diseases.

Disclosures: P.M. Miguel: None. **B. Barth:** None. **M.B. Alves:** None. **R. Dalle Molle:** None. **A. Gómez-Ilescas:** None. **A. Batra:** None. **D.M. Arcego:** None. **T. Zhang:** None. **X. Wen:** None. **C. Parent:** None. **N. O'Toole:** None. **M.J. Meaney:** None. **P.P. Silveira:** None.

Poster

PSTR160. Stress-Modulated Pathways: Social Stress

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR160.01/II9

Topic: F.03. Stress and the Brain

Support: NSF GRFP

Title: The BNST to NAc: a neural circuit for isolation induced social anxiety

Authors: *J. GRAMMER[1], M. ZELIKOWSKY[2], A. BOWLES[1], R. VALLES[1];
[2]Univ. of Utah, [1]Univ. of Utah, Salt Lake City, UT

Abstract: Prolonged social isolation has been shown to promote a host of deleterious effects on behavior including increased social anxiety; however, the neural mechanisms underlying this effect remains unknown. The nucleus accumbens (NAc) is integral to social behavior and has been implicated in social approach and avoidance. Alternatively, the dorsal bed nucleus of the stria terminalis (dBNST) is known for its role in mediating anxiety and social isolation stress. Thus, these regions represent prime candidates for the control of isolation-induced social anxiety. Importantly, the coordinated activity of these regions via their reciprocal, monosynaptic projections remains relatively unexplored on both an anatomical and functional level. Using fluorescent *in situ* hybridization (RNAscope) probing for the bi-directional neural activity marker EGR1, we found that isolation increases activity in the dBNST and decreases NAc activity. This suggests that an imbalance of activity in this pathway may underlie the effects of social isolation to impact social behavior. Next, using a chemogenetic loss-of-function approach, we found that the NAc is required for social approach in a three-chamber sociability assay. To comprehensively investigate social anxiety-like behavior in mice, we developed a new assay, Selective Access to Unrestricted Social Interaction (SAUSI), in which mice have the choice to pass through a small, one-way tunnel to freely interact with a conspecific mouse. Using in-depth observer scoring as well as automated machine-learning approaches, we found that social isolation stress increases social anxiety as indexed by social freezing and approach hesitancy, while altering complex social behaviors measured during free interaction. Collectively, these findings support the hypothesis that this pathway lies at the intersection between social behavior and anxiety. We are currently investigating the contribution of genetically defined dBNST to NAc projection cells in isolation-induced social anxiety-like behavior. Ultimately, our project defines a mouse model of social anxiety using a novel social choice assay that allows for motivated *and* free interaction, unveils isolation induced changes in neural activity in the dBNST and NAc, and explores a novel role for this circuit in isolation induced social anxiety-like behavior.

Disclosures: J. Grammer: None. **M. Zelikowsky:** None. **A. Bowles:** None. **R. Valles:** None.

Poster

PSTR160. Stress-Modulated Pathways: Social Stress

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR160.02/II10

Topic: F.03. Stress and the Brain

Support: NIH Grant 1R01MH132822-01

Title: Footshock-induced changes in social behavior are mediated by genetically-defined cells in the dBNST

Authors: *M. CONOSCENTI, N. POLL, L. MALADY, M. ZELIKOWSKY;
Univ. of Utah, Salt Lake City, UT

Abstract: The dorsal bed nucleus of the stria terminalis (dBNST) is an anatomically and functionally diverse brain region that mediates the behavioral and biological changes induced by a variety of stressors, including exposure to unpredictable footshock. Exposure to an acute stressor leads to widespread upregulation of corticotropin-releasing hormone (CRH), which in turn mediates stress-induced anxiety and aggression. In addition, we have found that social stress increases expression of tachykinin 2 (Tac2) in the dBNST, which is required for stress-induced persistent fear. Using *in situ* hybridization, we found that approximately 50% of Tac2-containing dBNST cells co-express CRH. However, the functional role of this unique overlapping population remains entirely unexplored. Here, we test the hypothesis that footshock-induced changes in social behavior are mediated by Tac2 and CRH co-expressing neurons in the dBNST ($dBNST^{Tac2 \cap CRH}$). Tac2-Cre, CRH-Cre, or Tac2-Cre;CRH-Flp transgenic C57Bl6/N mice were exposed to an acute footshock stressor (FS; 10, 1mA shocks randomly distributed across a 60-minute session), or the context without shock, and later tested for an array of social behaviors, including aggression, social interaction, anti-social behavior, and non-social behaviors using the resident intruder assay. We define the contributions of Tac2+, CRH+, and Tac2∩CRH+ neurons in the dBNST toward stress-induced social changes using a variety of approaches including chemogenetic and optogenetic manipulations, *in situ hybridization* using RNAScope, and *in vivo* calcium imaging using microendoscopes. Specifically, we found that FS-induced aggression and/or social avoidance are reduced when Tac2 and CRH+ dBNST neurons are silenced. Conversely, unstressed mice exhibit aggression and/or social avoidance when these cells are optogenetically stimulated. Finally, $dBNST^{Tac2 \cap CRH}$ seem preferentially active during a social interaction following exposure for footshock. Taken together, our findings illustrate a critical role for $dBNST^{Tac2 \cap CRH}$ neurons in social avoidance and aggression induced by stress.

Disclosures: M. Conoscenti: None. **N. Poll:** None. **L. Malady:** None. **M. Zelikowsky:** None.

Poster

PSTR160. Stress-Modulated Pathways: Social Stress

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR160.03/II11

Topic: F.03. Stress and the Brain

Support: NIMH Grant # MH132822
Whitehall Foundation
University of Utah Undergraduate Research Office
Sloan Foundation
Klingenstein Simons Foundation

Title: The lateral preoptic area controls the effects of social isolation on mouse courtship behavior and song

Authors: *E. CARROLL[1], J. LOVE[1], M. CONOSCENTI[1], A. COVINGTON[2], R. HANSON[4], M. ZELIKOWSKY[3];
[1]Univ. of Utah, Salt Lake City, UT; [2]Univ. of Utah, Kaysville, UT; [3]Dept. of Neurobio., Univ. of Utah, Salt Lake City, UT; [4]Blackrock Neurotech, Salt Lake City, UT

Abstract: Social communication is a vital component of courtship behavior across species. Prolonged periods of social isolation have been shown to impact mouse courtship behavior, however, the impact of social isolation to alter mouse courtship-related vocalizations during social interactions has been relatively unexplored. Through a series of behavioral experiments and acoustic recordings, we have discovered that chronic social isolation (SI) significantly alters mouse mating behavior and associated ultrasonic vocalizations (USVs). Detailed analysis of audio recordings collected from male mice during a homecage mating assay with a female conspecific reveal significant differences in USV production patterns, where isolated mice produce longer song syllables at a lower frequency with a smaller frequency range and fewer frequency jumps than group housed (GH) males. Importantly, we found that isolation negatively impacted mouse behavioral chains, altering the pattern of events comprising courtship chains in SI but not GH males. Moreover, we found a strong correlation between female defensive behaviors and male mounting attempts for GH, but not SI mice, suggesting that isolation disrupts the reciprocal balance between male-female behaviors during courtship. We complemented these behavioral and acoustic approaches with neural circuit analyses. Isolated males with altered USVs showed increased cfos expression in the lateral preoptic nucleus (LPO), a region previously implicated in mouse USVs more generally. To further probe the role of the LPO in SI mouse song, mice were injected with fluorescently encoded calcium indicators and implanted with GRIN lenses snapped to miniature microendoscopes to assess the in vivo activity of neurons in the LPO during mating behavior and USVs. Initial results revealed selective activity of neural ensembles in the LPO during SI-USVs. Collectively, our findings indicate that SI negatively modulates mouse courtship behaviors and song, and that this modulation is encoded by unique ensembles of neurons in the LPO.

Disclosures: E. Carroll: None. **J. Love:** None. **M. Conoscenti:** None. **A. Covington:** None. **R. Hanson:** None. **M. Zelikowsky:** None.

Poster

PSTR160. Stress-Modulated Pathways: Social Stress

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR160.04/II12

Topic: F.03. Stress and the Brain

Support: NIH Grant MH132822

Title: The Role of the Ventral Hippocampus in Trauma-Altered Sociability

Authors: *K. SATTLER, A. HEDGES, R. MILLER, M. ZELIKOWSKY;
Univ. of Utah, South Salt Lake, UT

Abstract: A single, acute traumatic experience can result in a host of negative behavioral effects, such as increased aggression, reduced sociability, and exaggerated fear responses to mild stressors. Despite the large body of research on the neurobiology of trauma, we nevertheless have a poor understanding of how the brain encodes complex trauma-associated changes in social behavior. The ventral hippocampus (VH) is well-suited for processing and motivating the multi-modal effects of trauma, as it receives inputs from sensory integration sites and sends output to regions involved in social and emotional behavior. Importantly, the VH projects to the basolateral amygdala (BLA), a central node for fear, the ventromedial hypothalamus (VMH), implicated in aggression, and the prefrontal cortex (PFC), known to be involved in social processing. Thus, we hypothesized that the effects of trauma to alter fear, aggression, and sociability are controlled by projection neurons in the VH. To test this hypothesis, male and female mice were injected with a virus encoding hM4D inhibitory DREADD into the VH to allow for chemogenetic silencing of the VH. They were then subjected to trauma (10 1mA foot-shocks over 60 minutes) or no trauma (60 minutes of context exposure) (off-drug), and subsequently tested for aggression, sociability, and social preference using the Resident Intruder assay (cohort 1) and the 3-Chamber Sociability assay (cohort 2). The DREADDs-activating ligand DCZ, or the control vehicle, was injected into half the mice before each assay using a counterbalanced design. We found that DCZ-treated hM4D mice showed attenuated trauma-induced aggression and trauma-reduced sociability compared to control mice. Next, we tested the hypothesis that distinct trauma-altered behaviors are controlled by overlapping populations of neurons on the VH using retrograde viral tracing, activity-dependent ensemble tagging, and immunohistochemistry. Collectively, our findings suggest a role for the VH as a central hub underlying trauma-altered sociability and provide insight into how experiencing a traumatic event can lead to diverse behavioral changes that extend beyond those classically studied.

Disclosures: K. Sattler: None. **A. Hedges:** None. **R. Miller:** None. **M. Zelikowsky:** None.

Poster

PSTR160. Stress-Modulated Pathways: Social Stress

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR160.05/II13

Topic: F.03. Stress and the Brain

Support: NIH Grant 1F31MH131359-01
NIH Grant MH132822

Title: The mPFC Tachykinin-2 system exerts top-down control over isolation-induced aggression.

Authors: *R. E. GATLIN[1], J. GAGON[1], H. WALKER[2], M. FLUCK[1], N. FROST[2], M. ZELIKOWSKY[1];
[1]Neurobio., [2]Neurol., Univ. of Utah, Salt Lake City, UT

Abstract: Chronically isolated mice show altered behaviors, including increased aggression, however the neural circuitry underlying this aggression is poorly understood. The medial prefrontal cortex (mPFC) is strongly involved in social behaviors, including aggression, however much of this work lacks cell-type specificity. Here, we propose a role for the mPFC Tachykinin-2 system in modulating social isolation-induced (SI) aggression. We previously established a role for the Tachykinin 2 (Tac2) neuropeptide system in SI alterations in behavior. Tac2 encodes the neuropeptide Neurokinin B, which binds the neurokinin-3 receptor (Nk3R), and while both are expressed in the mPFC, there has been no work performed to understand the role of this neuropeptide system in the mPFC in behavior. Using RNAscope and scRNAseq, we establish that cortical Tac2+ are a unique class of GABAergic neurons, whereas Nk3R+ neurons are glutamatergic pyramidal neurons. Given our previous work suggesting that isolation increases the activity of Tac2 neurons, we propose that the activity of the mPFC Tac2 system is increased by social isolation, preventing the mPFC from exerting top-down control over aggression. This is supported by literature establishing that reduced activity of mPFC pyramidal neurons corresponds with increased aggression. Thus, to understand the role of the mPFC Tachykinin-2 system in SI aggression, we used a viral-mediated chemogenetic approach to manipulate the activity of the mPFC Tachykinin-2 system to understand whether inhibition of Tac2 neurons or activation of Nk3R neurons was sufficient to reduce aggression. To this end, male and female Tac2-Cre mice were stereotaxically injected with either a Cre-dependent hM4D DREADD fused to mCherry or a Cre-dependent mCherry virus in the mPFC. In compliment, Nk3R-Cre mice were infused with a Cre-dependent hM3D DREADD or Cre-dependent mCherry virus. Following surgeries, animals were single housed for 4 weeks, then tested on the Resident Intruder assay twice to measure aggression. Ten minutes before the assay, mice were given an i.p. injection of either the DREADD-ligand, Deschloroclozapine (DCZ; 1μg/g), or vehicle. On the second test day, 48hrs later, the animals received the opposite solution. Aggression (number of attack bouts and time engaged in attack) towards an age and sex-matched Balb/c mouse was scored by a blinded experimenter. Both DREADD-mediated inhibition of mPFC Tac2+ neurons or activation of Nk3R+ neurons significantly reduced both the time engaged in attack and the number of attack bouts. These findings highlight a previously unknown role for Tac2/Nk3R signaling in mPFC in the control of SI aggression.

Disclosures: **R.E. Gatlin:** None. **J. Gagon:** None. **H. Walker:** None. **M. Fluck:** None. **N. Frost:** None. **M. Zelikowsky:** None.

Poster

PSTR160. Stress-Modulated Pathways: Social Stress

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR160.06/II14

Topic: F.03. Stress and the Brain

Support: NIMH MH132822

Title: Chronic social instability alters social and non-social behavior: a new role for oxytocin

Authors: *D. TAYLOR, M. A. CONOSCENTI, M. ZELIKOWSKY;
Neurobio., Univ. of Utah, Salt Lake City, UT

Abstract: Disruptions to social stability, including divorce, entrance into the foster care system, and, most recently, repeated COVID-associated school closures, represents a profound source of chronic stress. Importantly, social instability (SIN) has been shown to have long-lasting, negative effects on both socioemotional and physical health, nevertheless, the neurobiology underlying chronic social instability remains largely unknown. We developed a 21-day social instability protocol in mice to engender a state of social instability and examine the impacts on behavior and the brain. Following SIN, we evaluated social behavior using a resident intruder assay and fear learning and memory using a contextual fear conditioning task. We found that SIN animals display distinct social deficits characterized by a decrease in social interaction and an increase in asocial behaviors. Additionally, we found that SIN animals show a deficit in within-session extinction, suggesting that the SIN experience produces fear learning that is relatively resistant to extinction. Next, we examined the biological basis of SIN-altered social and fear behavior. Oxytocin, classically known for its role in maternity and pair bonding, has been recently implicated in non-traditional functions such as the formation of platonic relationships and same-sex social interactions. Thus, we hypothesized it could be critical for the formation of the state produced by chronic SIN. When we chronically blunted oxytocin signaling in SIN animals, using a systemic administration of an oxytocin receptor antagonist, we were able to reverse SIN-altered behaviors, suggesting that oxytocin signaling plays a key role in the expression of SIN on behavior. The dorsal CA2 region of the hippocampus (dCA2), a region known for its involvement in the formation of social memories and social interaction, and the central nucleus of the amygdala (CeA), a hub for the integration of emotional valence and memory, are both enriched with oxytocin-receptor positive neurons. Our ability to specifically reverse the social and fear learning phenotypes via oxytocin receptor antagonism suggests that these two structures may be sensitive to SIN-related changes in oxytocin signaling. As such, we are currently determining the region-specific function of oxytocin within the dCA2 and CeA to control SIN-altered social behavior and persistent fear in these regions, respectively. This work closes a

critical gap in our understanding of the behavioral and neurobiological impacts of SIN, identifying a novel, unexplored role for oxytocin signaling.

Disclosures: D. Taylor: None. **M.A. Conoscenti:** None. **M. Zelikowsky:** None.

Poster

PSTR160. Stress-Modulated Pathways: Social Stress

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR160.07/II15

Topic: F.03. Stress and the Brain

Support: R01MH051399
R01MH129306
NIH T32-MH087004
the Hope for Depression Research Foundation

Title: Resilient Specific Sex-Conserved Transcriptomic Changes in the Nucleus Accumbens Following Chronic Social Defeat Stress in Mice

Authors: *T. GYLES[1], E. M. PARISE[1], L. HOLT[1], A. MINIER-TORIBIO[1], T. MARKOVIC[1], L. PARISE[1], R. DURAND[1], L. LI[1], Y. YIM[1], C. BROWNE[1], A. GODINO[1], A. M. CARDONA-ACOSTA[3], M. ESTILL[1], C. A. BOLANOS-GUZMAN[4], S. J. RUSSO[5], E. J. NESTLER[2]; [1]Nash Family Dept. of Neurosci. and Friedman Brain Inst., [2]Icahn Sch. of Med. At Mount Sinai, Icahn Sch. of Med. At Mount Sinai, New York, NY; [4]Texas A&M Univ., [3]Texas A&M Univ., College Station, TX; [5]Icahn Sch. of Med. at Mount Sinai, Icahn Sch. of Med. at Mount Sinai, New York, NY

Abstract: Major depressive disorder (MDD) is a leading cause of disability and a leading contributor to suicide according to the World Health Organization. Chronic stress is a primary risk factor for MDD and is modeled in rodents using the chronic social defeat stress (CSDS) paradigm. Importantly, this paradigm allows for identifying animals across a continuum of responses from those that develop depression-like behavioral abnormalities, termed susceptible, to those that maintain mostly normal behavioral function, termed resilient. This approach has proven to be highly useful but has been mostly examined in male mice, leaving the examination of female mice understudied. Given that depression is more prevalent in women, it is crucial to investigate potential sex-specific molecular mechanisms underlying susceptibility vs. resilience. To address this gap, we conducted RNA-sequencing on female mice subjected to an adapted model of CSDS and identified transcriptional changes associated with the susceptibility-resilience spectrum across multiple brain regions.Initial comparison of this new dataset with published findings on male mice replicated earlier findings of striking sexual dimorphism in adaptations associated with susceptibility or with resilience in female vs. male mice in the brain regions studied. Despite this dimorphism, we identified a cluster of genes uniquely upregulated in the NAc of resilient female mice that overlapped ~50% with a previously identified resilient-

specific gene network in NAc of male mice. Within this gene network, two key driver genes, *Gprin1* and *Stx1a*, were predicted to regulate other genes in the network. To elucidate the role of these key driver genes, we are currently investigating the consequences of viral manipulation within medium spiny neuron subtypes of the NAc in both male and female mice prior to CSDS. Bilateral overexpression of GPRIN1 or STX1a in all NAc neurons of male mice induces a pro-resilient effect. Ongoing research aims to characterize the effects of cell-type-specific manipulation in both sexes and to examine changes in neuronal and circuit function that underlie the promotion of behavioral resilience.This study provides novel insights into the molecular mechanisms underlying stress resilience and offers valuable guidance for future efforts in antidepressant drug discovery. Our findings highlight the importance of considering sex-specific factors in understanding depression and developing targeted interventions.

Disclosures: **T. Gyles:** None. **E.M. Parise:** None. **L. Holt:** None. **A. Minier-toribio:** None. **T. Markovic:** None. **L. Parise:** None. **R. Durand:** None. **L. Li:** None. **Y. Yim:** None. **C. Browne:** None. **A. Godino:** None. **A.M. Cardona-Acosta:** None. **M. Estill:** None. **C.A. Bolanos-Guzman:** None. **S.J. Russo:** None. **E.J. Nestler:** None.

Poster

PSTR160. Stress-Modulated Pathways: Social Stress

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR160.08/II16

Topic: F.03. Stress and the Brain

Support: Center for Health and Wellbeing Grant, Princeton University

Title: Social isolation stress alters whole brain transcriptomics and translationally-relevant behaviors in a sexually dimorphic manner in Drosophila melanogaster

Authors: *E. TAWA[1], L. SCHNEPER[2], K. CHO[2], G. HILSCHER[2], D. NOTTERMAN[2]; [1]Neurosci., [2]Mol. Biol., Princeton Univ., Princeton, NJ

Abstract: Stress is known to contribute to an array of detrimental health effects, but our understanding of the mechanisms through which stress causes transcriptome-level changes in the brain is incomplete. *Drosophila melanogaster* provides a simple yet elegant model system to elucidate how environmental stress induces such changes and how these could be associated with specific behavioral responses. Researchers have begun to explore social isolation and overcrowding as stress paradigms in flies. However, it is unclear whether gene expression changes in the brain are common across these forms of environmental stress, the extent to which behaviors are altered by crowding and isolation, and how gene expression and behavior are differentially altered in males and females in response to stress. In this study, we aim to determine whether changes in gene expression induced by stress can be causally linked to changes in locomotor and social behaviors. We have exposed age-matched, male and female flies to chronic stress through either adult crowding or social isolation. We have shown that

social isolation stress alters gene expression in a sexually dimorphic manner in candidate fly anxiety genes, with elevated expression fold change in *5-HT2B* (mean 1.69, SD 0.60) and *CCKLR-17D1* (mean 1.79, SD 0.61) in males but not females. We have used ORE-R flies and CRISPR mutants of the cholecystokinin-like receptor genes (*CCKLR-17D1* and *CCKLR-17D3*) to show that social isolation induces hyperactivity in wild type males ($p<0.0001$, Mann-Whitney Test) but not females ($p=0.2127$, Mann-Whitney Test), yet this behavioral difference is abrogated upon knockout of these 2 candidate genes. Furthermore, we have suggestive evidence that females have a lower preference for sucrose compared to males following social isolation ($p=0.0485$), indicative of anhedonia-like behavior. We have also carried out RNA-seq and found that genes implicated in networks for locomotion, memory, brain development, cellular response to dopamine, and positive regulation of feeding behavior are regulated in opposite directions between males and females following social isolation and overcrowding. Our findings provide transcriptomic and behavioral evidence that differences exist in how social stress impacts male and female flies. Future directions include identifying a specific network implicated in stress-relevant behaviors and testing knock out and overexpression models to establish causal relationships. Together, these findings could help identify novel targets for sex-specific therapeutic interventions to alleviate the psychiatric symptoms of stress-related disorders.

Disclosures: E. Tawa: None. **L. Schneper:** None. **K. Cho:** None. **G. Hilscher:** None. **D. Notterman:** None.

Poster

PSTR160. Stress-Modulated Pathways: Social Stress

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR160.09/II17

Topic: F.03. Stress and the Brain

Support: 5R01MH111918

Title: Variation in Amygdala Volume Impacts Endophenotypes of Resilience and Susceptibility using Chronic Social Defeat Stress in Male and Female Mice

Authors: *H. MCBRIDE[1], D. FORTUNA[2], L. BRYAN[4], K. R. ANDERSON[5], A. MANGANARO[3], D. DUMITRIU[6];
[1]NYSPI, New York, NY; [2]Neurosci., [3]New York State Psychiatric Inst., New York, NY; [4]Columbia Univ., New York, NY; [5]Natl. Univ. of Ireland, Galway, Ireland; [6]Columbia Univ. Irving Med. Ctr., New York, NY

Abstract: Chronic stress exposure has a significant impact on disease burden worldwide. Stressful life events can impact neurology to induce mental disorders and lead to increased rates of chronic mental health diseases. Understanding the underlying neuronal and physical structures that might make subjects more susceptible to stress can help determine the risk for chronic mental health disorders. The amygdala is a brain structure that has long been associated with

emotion processing and risk-taking behaviors. Subjects with abnormalities in amygdala function and size may be predisposed to mental health disorders. The rodent model Chronic Social Defeat Stress (CSDS) is used to uncover the impact of a stressor on brain function and structure. Social defeat stress is a well-validated model of divergent stress response within rodent homogenous populations, allowing for categorization into “resilient/approach” and “susceptible/avoidant” subpopulations. CSDS exposes mice to repeat bouts of social aggression by an aggressive CD1 mouse over a ten-day period, which leads to social avoidance (CSDS susceptible) and normal social approach (CSDS resilient) phenotypes. Here, we use CSDS to determine if resilience and susceptibility are predisposed by alterations in brain structure. Specifically, we hypothesize that a mouse with a larger amygdala will show more risk-taking and emotional processing behavior, and thus will be more likely to have a higher SI ratio and approach phenotype. Additionally, we hypothesize that divergent phenotypic stress-responses are conferred by underlying variability in functional connectivity prior to stress as well as strengthened stress-activation circuits during chronic stress exposure leading to maladaptive behaviors. The population undergoing CSDS showed a resilient phenotype in 40% of females and males. Preliminary data (n=10), shows a positive correlation between SI score and amygdala size as a proportion of total brain area (R^2= 0.6802). Additional cohorts are needed to confirm these results, but early data suggest larger amygdala volume may be correlated with increased resilient phenotype. Further research will be conducted to determine the neural substrates within the amygdala of these endophenotypes.

Disclosures: H. McBride: None. **D. Fortuna:** None. **L. Bryan:** None. **K.R. Anderson:** None. **A. Manganaro:** None. **D. Dumitriu:** None.

Poster

PSTR160. Stress-Modulated Pathways: Social Stress

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR160.10/II18

Topic: F.03. Stress and the Brain

Support: IN208722

Title: Effects of Social Isolation and Loneliness during the COVID-19 Pandemic: Insights from an Animal Model

Authors: A. C. ARMAS SANCHEZ, P. TORRES-CARRILLO, Y. B. VIDAL DE LA O, D. B. PAZ-TREJO, ***H. SANCHEZ-CASTILLO**;
Univ. Nacional Autonoma de Mexico, Mexico City, Mexico

Abstract: The global spread of the severe acute respiratory syndrome coronavirus 2 (SARS-CoV-2), the virus responsible for COVID-19, has had a significant impact in our society and has led to the implementation of measures to prevent massive infections and reduce the burden on healthcare systems worldwide. Two of the most important adopted measures were the social distancing and voluntary home isolation, recommended by worldwide organizations such as the

World Health Organization. However, the non-regulated social distancing and social isolation has also raised concerns about their potential repercussions on people's mental health. Loneliness and social isolation have been identified as important risk factors that can have adverse consequences for health. Several studies have been shown an association between loneliness and social isolation and negative health outcomes. The main objective of this work was to describe and standardize an animal model of social isolation stress (AIS-ST-I) that would serve for the evaluation of behavioral and physiological effects of social isolation in male and female rats. The rats were exposed to a 60-day period of social isolation (starting from postnatal day 30). During this time, measurements of corticosterone (CORT) levels were taken. Additionally, behavioral tests were conducted using the Barnes Maze. The obtained results indicated that prolonged social isolation had a significant impact on the rats. There was an increase in corticosterone levels compared to the control group, and statistically significant differences were observed based on the stress treatments, suggesting that stress induced by social isolation activated the physiological stress response in the animals. Furthermore, treatment-dependent behavioral alterations were found in the conducted tests.

Disclosures: A.C. Armas Sanchez: None. **P. Torres-Carrillo:** None. **Y.B. Vidal de la O:** None. **D.B. Paz-Trejo:** None. **H. Sanchez-Castillo:** None.

Poster

PSTR161. Sleep: Relationship to Disease, Disorders, Cognition, and Behavior

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR161.01/II19

Topic: F.07. Biological Rhythms and Sleep

Title: Amyotrophic Lateral Sclerosis (ALS) Disrupts Sleep Architecture and Reduces Rapid Eye Movement Sleep (REM) and Slow-Wave Sleep (SWS)

Authors: *S. S. ALDALIL[1], T. KJÆR[2];
[1]Dept. of Neurosci., Brandeis Univ., Waltham, MA; [2]Neurol., Neurol. Dept. at Zealand Univ. Hosp., Roskilde, Denmark

Abstract: Amyotrophic lateral sclerosis (ALS) is the most common and severe form of motor neuron disease (MND) that ultimately leads to generalized muscle weakness, respiratory failure, and eventually death. One of the hallmarks of ALS is the neurodegeneration of motor neurons in the brain and spinal cord. Sleep disturbances are common symptoms in patients with ALS and significantly add to the burden of the disease for patients and caregivers alike. Disruption of sleep can be caused by physical symptoms, such as muscle cramps, pain, reduced mobility, mucus retention, and restless legs syndrome which are all characteristic of ALS. In particular, sleep-disordered breathing (SDB) and nocturnal hypoventilation (NH) are common among ALS patients. In the present study, we examined sleep architecture and its objective quality, using polysomnography (PSG) and sleep scoring, in an ALS patient and age-matched healthy control. Moreover, a follow-up PSG recording was conducted 3 months after the first PSG recording to

investigate whether there will be further disruption in sleep architecture and deterioration in its objective quality over the course of the disease. We found reductions in total sleep time (TST), REM latency, and duration in the ALS patient when compared to the healthy control. Additionally, we found decreases in N2, and slow wave sleep (SWS) as well as REM sleep in the ALS patient 3 months following the first PSG recording. On the other hand, increases in arousal-awakening index (AAI), and N1 sleep were observed when the two PSGs were compared. Taken together, these data provide a more quantitative and qualitative assessment of ALS-induced sleep disturbances as well as what certain sleep stages can be most vulnerable in ALS.

Disclosures: S.S. Aldalil: None. **T. Kjær:** None.

Poster

PSTR161. Sleep: Relationship to Disease, Disorders, Cognition, and Behavior

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR161.02/II20

Topic: F.07. Biological Rhythms and Sleep

Support: Congressionally Directed Medical Research Programs (CDMRP)

Title: Beyond Counting Sheep: The Interplay Between Sleep and Cognitive Performance in Military Service Members With Mild Traumatic Brain Injuries

Authors: *H. RIZEQ[1,2], C. DAQUINO[1,2], W. ZHENG[1,2], M. L. ETTENHOFER[3,4,5,6], S. GIMBLE[3,4,5], L. HUNGERFORD[3,4,5], E. CHINOY[1], P. SESSOMS[1];
[1]Naval Hlth. Res. Ctr., San Diego, CA; [2]Leidos, Inc., Reston, VA; [3]Naval Med. Center, San Diego, San Diego, CA; [4]Traumatic Brain Injury Ctr. of Excellence, Silver Spring, MD; [5]Gen. Dynamics Information Technol., Falls Church, VA; [6]UCSD, La Jolla, CA

Abstract: Individuals with mild traumatic brain injury (mTBI) often experience sleep disturbances and report cognitive dysfunction. Existing literature has primarily focused on either sleep disturbances or cognitive performance in mTBI, with limited research examining the effects on military related performance tasks. To address this gap, 15 active duty military members (10 mTBI and 5 controls) were recruited for this study. Participants performed two one-back tests in an immersive virtual reality environment. Simulating a military patrol mission, the tasks required participants to recognize, aim, and shoot at an avatar appearing on the screen, shooting either at the upper or lower part of the avatar, depending on whether the current avatar was the same as or different from the previous one (one-back). In the second version of the task, battlefield radio chatter was continuously broadcast to enhance the effect of realism of the patrol mission and to increase the cognitive load by requiring the participant to orally respond to specific chatter cues. Sleep was assessed using the ReadiBand® sleep tracker (Fatigue Science, Vancouver, BC), capturing two variables: wake after sleep onset (WASO) and total sleep time (TST). Given the limited sample size, only descriptive results of data analysis were reported. The control group outperformed the mTBI group in the shooting task. The mTBI participants

responded to targets more slowly than the controls (shooting response time: 1335.8 ms ± 86.2 ms vs 1287.4 ms ± 135.7 ms). The mTBI group made more incorrect target shots than the controls (percent correct shots: 85.3% ± 7.3% vs 90.6% ± 5.5%). Similarly, the mTBI participants failed to respond to chatter cues more often than the controls (percent correct responses: 85.67% ± 8.1% vs 88.7% ± 7.7%). Interestingly, and contrary to expectations, controls demonstrated slightly poorer sleep compared with the mTBI group on the night prior to testing. Controls had a mean WASO of 54 ± 33 minutes, while the mTBI group exhibited a mean of 46 ± 42 minutes. For TST, controls had a mean of 376 ± 102 minutes, and the mTBI group had a mean of 384 ± 58 minutes. While these preliminary findings suggest that prior-night sleep may not reliably predict next-day performance among individuals with mTBI and controls, it is important to note that these conclusions are drawn from a limited sample size. Ongoing data collection and analyses with larger sample sizes and assessing sleep metrics over a longer period will help inform this potentially unexpected disparity. Generally, there is a need for further investigation into the complex interplay between sleep, mTBI pathology, and cognitive functioning.

Disclosures: H. Rizeq: None. **C. Daquino:** None. **W. Zheng:** None. **M.L. Ettenhofer:** None. **S. Gimble:** None. **L. Hungerford:** None. **E. Chinoy:** None. **P. Sessoms:** None.

Poster

PSTR161. Sleep: Relationship to Disease, Disorders, Cognition, and Behavior

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR161.03/II21

Topic: F.07. Biological Rhythms and Sleep

Title: Investigation of potential links between neural and galvanic skin response dynamics during sleep

Authors: *J. A. DECKER[1], S. VIJAYAN[2];
[1]Sch. of Biomed. Engin. and Sci., [2]Sch. of Neurosci., Virginia Tech., Blacksburg, VA

Abstract: Emotional memory consolidation (EMC) is a process critical to emotional memory processing, occurring during sleep. Disruptions to EMC can be caused by disorders such as post-traumatic stress disorder (PTSD), which affect a large segment of the population. The galvanic skin response (GSR) offers the potential for a lightweight method of tracking emotional activity during sleep, as it is a marker of emotional arousal in waking subjects (Bouscein et al, 2012). GSR storms, a unique GSR event that occurs throughout sleep, is of particular interest, as it may be a marker of increased emotional activity (Sano et al, 2014). We investigate the potential links between neural dynamics and GSR storms during sleep. Using a combination of standard polysomnography and wearable devices to record GSR, we recorded sleep data in nap and overnight sessions in healthy 18-26-year-old participants, and analyzed the spectral and temporal relationship between the data (N=5). An initial investigation revealed both a temporal and spectral relationship between neural and GSR dynamics. In N1 and N2 sleep, the temporal relationship between the two signals is similar to the one seen in wake, but in N3 sleep, a lag

between the neural dynamics and the GSR response can be seen (Bouscein et al, 2012). The spectral relationship shows sleep stage specific differences in power, particularly in the high alpha/low beta frequency band. In N2 sleep, this relationship is most visible, as the peak seen in this range shifts dependent upon location. In other sleep stages, the alpha peak is less prominent or not present. These findings indicate a potential relationship between neural and GSR dynamics during sleep, which could be studied further to develop a lightweight method of detecting specific neural dynamic patterns. These relationships could be informative in studying memory encoding and recall, as well as a number of neurological disorders affecting sleep, like PTSD and depression.

Disclosures: J.A. Decker: None. **S. Vijayan:** None.

Poster

PSTR161. Sleep: Relationship to Disease, Disorders, Cognition, and Behavior

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR161.04/II22

Topic: F.07. Biological Rhythms and Sleep

Title: Sleep as a mechanism for asthma and anxiety

Authors: *R. KING[1], E. ROUSTON[1,2], R. GERARDO[3], R. HIGGINS[1], S. CAVIGELLI[1], W. HORTON[1];
[1]Pennsylvania State Univ., State College, PA; [2]Tufts, Medford, MA; [3]Cornell Univ., Ithaca, NY

Abstract: Worldwide, asthma affects over 300 million people of all ages, resulting in significant socio-economic losses (Giacco et al. 2016). With prevalence rates of approximately 9.5%, asthma is the leading chronic illness of children and adolescents, and has been correlated with a plethora of adverse outcomes, including an increase in learning difficulties, behavioral problems, and social problems (Dudeney et al., 2017). Notably, psychological disorders—particularly internalizing disorders like anxiety—are higher in asthmatic youth than healthy youth, with 22.7% and 6.9% prevalence rates, respectively (Dudeney et al., 2017; Lawrence et al., 2015). We hypothesized that sleep could be the mediating factor that links these two diseases, and performed a literature review to determine if extant literature supports this hypothesis. We identified 132 papers from multiple databases that address this link. In particular, we found a link between the severity of asthma and an increase in obstructive sleep apnea (OSA, a disease that affects sleep quality) risk, indicating a near 9-fold increased risk for the prevalence of OSA symptoms (Wang et al., 2017); and up to 50% of asthmatic patients have OSA. Additionally, we found an association between OSA and anxiety, with increased OSA symptoms contributing to increased symptoms of anxiety (Vanek et al., 2019). Thus, with data from multiple databases, it was found that sleep may be a mediator of this relationship, specifically, the development of sleep disordered breathing/obstructive sleep apnea.

Disclosures: **R. King:** None. **E. Rouston:** None. **R. Gerardo:** None. **R. Higgins:** None. **S. Cavigelli:** None. **W. Horton:** None.

Poster

PSTR161. Sleep: Relationship to Disease, Disorders, Cognition, and Behavior

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR161.05/II23

Topic: F.07. Biological Rhythms and Sleep

Title: Spatial heterogeneity of cerebrovascular response to sleep apnea: a pilot near-infrared spectroscopy (NIRS) study

Authors: A. ASGARLI, A. L. CONRAD, ***J. LIU**;
Univ. of Iowa, Iowa City, IA

Abstract: **Introduction:** Intermittent hypoxia (IH) occurs during sleep in obstructive sleep apnea (OSA), which is a very common disease. Cerebral blood flow (CBF) often increases in response to IH and may compensate for the detrimental effects of IH. If this CBF response is heterogeneous across the cerebral cortex, cortical regions with weaker CBF response may be more impaired by IH. Here we studied, for the first time, the spatial heterogeneity of CBF response to IH during sleep by using multi-channel near-infrared spectroscopy (NIRS) during overnight sleep in patients with OSA. **Methods:** We recruited 12 subjects previously diagnosed with a moderate or severe degree of OSA. Each subject took one night of natural sleep without any treatment for OSA, in a recliner in semi-upright position with a neck pillow so that the NIRS sources and detectors were not disturbed during sleep. NIRS was continuously recorded (sampling rate: 10.2 Hz), from 4 channels placed on scalp over the left angular gyrus, 4 over the right angular gyrus, 4 over the medial prefrontal cortex, and 4 over the right dorsolateral prefrontal cortex. Tissue oxygen saturation (StO_2), a metric highly correlated with CBF, was derived from the NIRS signals and compared across the 16 channels. Peripheral arterial tonometry and oxygen saturation (SaO_2) were simultaneously recorded using a medical-grade device that is clinically used to diagnose OSA. IH events were identified as transient decreases of SaO_2 by at least 4%. **Results:** NIRS signal quality was good in all channels in 11 of the 12 subjects, but IH events occurred frequently (more than 10 per hour) in only 6 of them, in part because of the semi-upright sleep position. Among these 6 subjects, two showed no significant difference in StO_2 across all channels, as well as strong correlation between StO_2 and SaO_2. The other four subjects showed significant differences in the StO_2 responses to IH events between left and right angular gyrus and/or between the prefrontal cortex and the angular gyrus. The main differences were that a few channels showed earlier and stronger increases of StO_2 than the rest of channels. **Discussion:** This pilot study illustrates that the spatial distribution of CBF response to IH during overnight natural sleep can be studied using the multi-channel NIRS, with advantages of high signal quality and comfort. Our key novel finding is that the CBF response to IH during sleep may be different across the cerebral cortex in some, but not all, OSA patients.

Future NIRS studies with whole-head coverage and many more subjects are needed to understand the functional significance of this novel finding of spatial heterogeneity.

Disclosures: **A. Asgarli:** None. **A.L. Conrad:** None. **J. Liu:** None.

Poster

PSTR161. Sleep: Relationship to Disease, Disorders, Cognition, and Behavior

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR161.06/II24

Topic: F.07. Biological Rhythms and Sleep

Support: NIGMS Award Number U54GM133807

Title: Exploring the impact of the "Night-float rotation" on medical residents: Effects on sleep/wake cycles and circadian control of temperature

Authors: *J. R. MARRERO[1], Y. DE JESÚS-BERRÍOS[2], A. C. SEGARRA[3], F. RAMIREZ-MARRERO[5], L. ORTA ANÉS[4], C. M. NAZARIO[4], J. L. AGOSTO[6];
[1]Biol., UPR - Rio Piedras, San Juan, PR; [2]UPR - Med. Sci. Campus, San Juan, PR; [3]Physiol., [4]Univ. of Puerto Rico Med. Sci. Campus, San Juan, PR; [6]Biol., [5]Univ. of Puerto Rico, Rio Piedras Campus, San Juan, PR

Abstract: Introduction: Sleep disruption during medical residency is a significant risk factor for medical trainees' exhaustion and can lead to burnout. This leads to an increased risk of medical errors, development of anxiety and emotional health complications like depression and ideation of self-harm. Individual coaching intervention with physicians in-training revealed that some residents struggle with sleep difficulties during the night-float rotation. We propose that differences in the synchrony of the residents' circadian oscillators and their work shift may be associated with sleep disturbances.**Methods**: This is a longitudinal cohort study of repeated measures in sixteen medical residents. Participants were followed for 3 consecutive rotation blocks covering a diurnal pre-float rotation, the night float rotation and a post-float diurnal rotation. Participants also completed a series of questionnaires through each rotation. A dermal sensor was used to evaluate the oscillations in core body temperature to determine circadian oscillation. A wrist sensor monitored sleep and activity of the participants to assess the degree of synchrony between parameters measured. Saliva samples were collected to measure cortisol and melatonin levels, to analyze stress and explore circadian fluctuations.**Analysis**: Information was collected for each participant to measure the outcome variation according to hazard exposures and other characteristics. We have taken repeated measurements on the same participants at various points in time to increase statistical power. Due to the small sample size, the importance of study's results is not based solely on statistical significance, but also on clinical significance. We used specialized software to study the oscillation of measurements, assess the shift in phases and periodicity along the night float rotation, and explore possible causal relationships between parameters measured.**Conclusions**: Evaluation of temperature data shows a disruption in the

oscillation of temperature including shifts in phase and period during the night float. Preliminary sleep analysis shows differences in sleep duration, onset, and timing.

Disclosures: **J.R. Marrero:** None. **Y. De Jesús-Berríos:** None. **A.C. Segarra:** None. **F. Ramirez-Marrero:** None. **L. Orta Anés:** None. **C.M. Nazario:** None. **J.L. Agosto:** None.

Poster

PSTR161. Sleep: Relationship to Disease, Disorders, Cognition, and Behavior

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR161.07/Web Only

Topic: F.07. Biological Rhythms and Sleep

Support: AIMSR/IRB/RC/2023/01/007

Title: Sleep quality and it's predictors among undergraduate medical students of a teaching institute in Telangana, South India : a cross-sectional study

Authors: *S. PATNALA[1], A. MOHANDAS[2];
[2]Community Med., [1]Apollo Inst. of Med. Sci. and Research, Hyderabad, Hyderabad, India

Abstract: Medical students are a vulnerable group prone to sleep disturbances. Along with academic pressure, they are expected to navigate life through duties that embrace long on-calls, emotional challenges and exhaustion related to witnessing human misery. Lack of adequate sleep can impair cognition and fine learning skills and cause deficits in attention, drowsiness while driving, risk-taking behavior, depression and impaired relationships. Hence, sleep is of particular interest in medical students due to its potential impact on learning the technical aspects of curriculum and providing quality patient care. A similar comparative study done among undergraduate medical students of Rwanda found a high prevalence of poor sleep quality, 80% was published in the Nature Journal. The present study was conducted to measure sleep quality and determine the factors associated with sleep quality among undergraduate medical students of a teaching institute in Telangana, South India. A cross-sectional study was conducted over 6 months among a calculated sample size of 401. A questionnaire including the socio-demographic details of the student, psychiatric and medication history, occurrence of headaches in the past month, night time screen use, screen time per week, perceived academic stress, self-satisfaction with performance, and the result of last exam was followed by Pittsburgh Sleep Quality Index which includes 11 questions assessing time of quality sleep, frequency of troubled sleep, requirement of medicines to induce sleep, etc over last 1 month and calculates the score using 7 components where a score of more than 5 in PSQI indicates poor sleep quality. Mean age of study participants was 20.41 years (SD=2.08). 3.2%had known psychiatric conditions like anxiety,depression. 4.2% had known comorbidities like Hypothyroidism, Asthma, PCOS, etc. It was found 70% of the students were found to have poor sleep quality. The mean global PSQI score among the students was 6.22(SD=2.9). Factors significantly associated with poor sleep quality were perceived academic stress [p value = 0.009; OR=2.154(CI=1.250-3.713)],

occurrence of headaches and use of self-medication[p value 0.008; OR= 1.868(CI=1.178-2.964)] and night time screen use[p value<0.001; OR=18.12(CI=5.48-59.87)]. Average screen time was noted as 5 hours 33 minutes(SD= 2 hours 38 minutes). These findings highlight that sleep deprivation and poor sleep quality is a major concern for physical & mental health and the importance of sleep hygiene, especially by reducing the usage of screens close to bedtime and addressing other health conditions and mental health issues to improve overall sleep quality and well-being.

Disclosures: S. Patnala: None. **A. Mohandas:** None.

Poster

PSTR161. Sleep: Relationship to Disease, Disorders, Cognition, and Behavior

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR161.08/II25

Topic: F.07. Biological Rhythms and Sleep

Support: Armenise-Harvard Foundation CDA
Wellcome Trust Seed Award (217546/A/19/Z)

Title: Adolescent chronic sleep restriction promotes alcohol drinking in adulthood: evidence from epidemiological and preclinical data

Authors: *O. FANIYAN[1], D. MARCOTULLI[5], R. SIMAYI[2], F. DEL GALLO[3], S. DE CARLO[3], E. FICIARÀ[3], D. CARAMASCHI[6], R. RICHMOND[7], D. FRANCHINI[8], M. BELLESI[8,4], R. CICCOCIOPPO[3], L. DE VIVO[3];
[2]Life and Hlth. Sci., [3]Sch. of Pharm., [4]Sch. of Biosci. and Vet. Med., [1]Univ. of Camerino, Camerino, Italy; [5]Sci. of Publ. Hlth. and Pediatrics, Univ. of Torino, Turin, Italy; [6]Psychology, Univ. of Exeter, Exeter, United Kingdom; [7]Bristol Population Hlth. Sci. Inst., [8]Pharmacol. and Neurosci., Univ. of Bristol, Bristol, United Kingdom

Abstract: Insufficient sleep is associated with the deterioration of cognitive functions, altered emotions, and an enhanced propensity to engage in risk-taking behaviours. In modern society, chronic sleep restriction (CSR), defined as gaining less than 8-10 hours of sleep *per* night for consecutive nights, has become epidemic among adolescents and has been proposed as a contributing factor to the development of alcohol use disorders (AUD) and related neuropsychiatric comorbidities later in life. However, a causal link between insufficient sleep and the development of AUD remains hazy. Here, we used epidemiological data and preclinical animal model to investigate the longitudinal causal link between adolescent CSR and alcohol use.The epidemiological dataset included 5497 participants (47.1% males) of the Avon Longitudinal Study of Parents And Children study. Sleep and alcohol drinking measures were collected from interviews and questionnaires at 15 and 24 years of age. Multivariable linear regressions and a cross-lagged autoregressive path model were used to test cross-sectional and longitudinal associations. Preclinical data were collected by sleep restricting male Marchigian

Sardinian alcohol preferring rats (N=40) from post-natal day 25 to 55 and measuring voluntary alcohol drinking concurrently and in adulthood. Behavioural tests were carried out 1 and 5 weeks after the end of the CSR protocol to assess anxiety, risky behaviour, and despair in the short- and long-term. Automatic CSR was validated by polysomnography in adolescent and young adult rats (n=9).After adjusting for covariates, cross-sectional associations between sleep insufficiency and alcohol consumption were observed at 15 years of age. The longitudinal analysis and the autoregressive cross-lagged path model highlighted that alcohol consumption at 24 years was predicted by insufficient sleep at 15 years whilst alcohol drinking at 15 years could not predict sleep problems at 24 years. In the preclinical model, CSR escalated alcohol consumption during adolescence and led to higher alcohol drinking and altered behavior later in adulthood, relative to controls. CSR rats showed increased risky behavior in the novelty-suppressed feeding test and increased total ambulatory time and distance travelled in the open field. No differences were found in the forced swim test or in the light/dark box test. In summary, our findings support the longitudinal and causal relationship between adolescent insufficient sleep and higher adult alcohol consumption, accompanied by long lasting altered behaviour.

Disclosures: O. Faniyan: None. **D. Marcotulli:** None. **R. Simayi:** None. **F. Del Gallo:** None. **S. De Carlo:** None. **E. Ficiarà:** None. **D. Caramaschi:** None. **R. Richmond:** None. **D. Franchini:** None. **M. Bellesi:** None. **R. Ciccocioppo:** None. **L. de Vivo:** None.

Poster

PSTR161. Sleep: Relationship to Disease, Disorders, Cognition, and Behavior

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR161.09/II26

Topic: F.07. Biological Rhythms and Sleep

Title: Chrono-nutrition Relationship timing food and BMI in a sample of children from Xalapa, Mexico

Authors: G. MARCIAL-HERNANDEZ[1], P. RENDON-GUTIERREZ[1], L. CABALLERO-TAPIA[2], P. GARCIA-ESQUIVEL[3], M. ZAMUDIO-AGUILAR[2], E. AGUILAR-SANCHEZ[2], M. ROSAS-NEXTICAPA[1], ***M. A. MELGAREJO-GUTIERREZ**[2];
[1]Facultad de Nutrición, [2]Med. Fac., Univ. Veracruzana, Veracruz, Mexico; [3]Programa Estatal de Estilos de Vida Saludable, Secretaria de Educacion de Veracruz, Veracruz, Mexico

Abstract: Circadian rhythms, metabolism, and nutrition are intimately linked, however; the effect of alimentation rhythm in a children population is poorly studied. Chrono-nutrition is an emerging field of study where there are three dimensions Timing, frequency and the regularity of the alimentation in order to keep a good metabolic health. In this way, several studies have been carried out to adults and night workers and through this we could figure out that putting out of order the time of our food and sleep can produce some metabolic illnesses, due to the desynchronization neuroendocrine of hormones which participate in the regularization of the appetite, satiation and other ones. It is important emphasize that the investigation in children´s

chrono-nutrition is not often evaluated because there is no conclusive information about the alimentation rhythm and metabolism. Therefore, the main objective of this study was identifying the food schedule of children (6-12 years) at the public schools in Xalapa, Veracruz. In order to have this information we applied a Chrono-nutrition profile which was adapted for schoolchildren and this one was previously authorized consent. The result showed that the weekday breakfast was at 07:41 ± 0.15 h in a total population. Boys have breakfast at 07:17 ± 0.18 h and girls have breakfast at 8:00 ± 0.26 h. A comparison analysis was performed between both groups, using the student's T test, Mann-Whitney U, in which it was found that there is a difference between the breakfast time of both groups P=0.021. The breakfast schedule on weekends was at 09:39 ± 0.19 hours in a total population. Boys have breakfast at 09:10 ± 0.27 hours and girls have breakfast at 10:12 ± 0.24 hours no statistically significant difference was found between the breakfast time of both groups P=0.104. On the other hand, in the anthropometric data of the studied population, it was found that the BMI is 17.2 ± 0.6 kg/m2 in a total population. There is a balance among the breakfast, lunch and dinner during the weekdays, however; on the weekends they had breakfast two hours later than on weekdays. There was no significant change regarding of the lunch and dinner considering the weekdays. An imbalance having breakfast some hours later than usually can provoke some hormonal and molecular changes in the long term, it can increase a risk in order to develop metabolic illnesses in adulthood, based on previous studies. It is necessary to make some analysis with more information about it in order to show a relation between these.

Disclosures: G. Marcial-Hernandez: None. **P. Rendon-Gutierrez:** None. **L. Caballero-Tapia:** None. **P. Garcia-Esquivel:** None. **M. Zamudio-Aguilar:** None. **E. Aguilar-Sanchez:** None. **M. Rosas-Nexticapa:** None. **M.A. Melgarejo-Gutierrez:** None.

Poster

PSTR161. Sleep: Relationship to Disease, Disorders, Cognition, and Behavior

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR161.10/II27

Topic: F.07. Biological Rhythms and Sleep

Support: The Hong Kong Research Grants Council Collaborative Research Fund (Ref: C7069-19G)
The Hong Kong Research Grants Council General Research Fund (Ref: 17600522)

Title: Moderating Effects of Sleep Quality on the Relationship between Multimodal Brain Networks and Glymphatic System in Older Adults

Authors: *J. MA, T. M. C. LEE;
the Univ. of Hong Kong, Hong Kong, China

Abstract: Introduction: Sleep complaints are prevalent among older adults, and poor sleep quality is a major risk factor for late-life mental health issues. Previous studies have demonstrated the substantial effects of sleep on structural/functional connectivities (SC/FC) in the human brain and the glymphatic system separately, with implications for mental health. However, the interplay between the sleep, SC/FC, and glymphatic system, and how this relationship contributes to mental health remain unclear. Therefore, we employed multimodal neuroimaging techniques to investigate the neurophysiological mechanisms underlying the association between sleep and mental health in older adults.
Methods: A cross-sectional study was conducted on a sample of 72 healthy older adults aged 60-92 years. The participants were divided into two groups based on their sleep quality: good sleepers and bad sleepers. Mental health status was assessed using the Hamilton Anxiety Rating Scale (HAM-A) and Hamilton Depression Rating Scale (HAM-D). Correlation between SC and FC was calculated to evaluate the SC-FC coupling, and the diffusion tensor imaging along the perivascular space (DTI-ALPS) index was calculated to measure the functioning of the glymphatic system.
Results: Significant group difference was observed in mental health, with bad sleepers having higher HAM-A and HAM-D scores than good sleepers ($ts > 2.045$, $ps < 0.045$). DTI-ALPS was negatively correlated with SC-FC coupling strength ($r = -0.251$, $p = 0.036$), suggesting more flexible neural communication in participants with better glymphatic function. However, this relationship was moderated by participants' sleep quality. The negative correlation between DTI-ALPS and SC-FC coupling was significant in good sleepers but not in bad sleepers. More specifically, the disrupted SC-FC coupling in the orbitofrontal area positively correlated with HAM-A and HAM-D scores ($rs > 0.313$, $ps < 0.008$).
Conclusion: Results of our study revealed a disrupted association between glymphatic function and SC-FC coupling in older adults with poor sleep, which further leads to worse anxious and depressive problems in this population. Overall, our findings reemphasize the importance of sleep for the mental health of older adults and advance the understanding of the interplay between sleep, glymphatic system, and brain health.

Disclosures: **J. Ma:** None. **T.M.C. Lee:** None.

Poster

PSTR161. Sleep: Relationship to Disease, Disorders, Cognition, and Behavior

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR161.11/II28

Topic: F.07. Biological Rhythms and Sleep

Support: NINDS Intramural Research Program

Title: Dream-report length is correlated with sleep functional correlations between posterior cingulate cortex and visual and memory areas

Authors: *H. HALIVNI, H. GURA, J. A. DE ZWART, H. MANDELKOW, F. N. YANG, P. VAN GELDEREN, J. H. DUYN, D. PICCHIONI;
NIH, Natl. Inst. of Neurolog. Disorders & Stroke (NINDS), Bethesda, MD

Abstract: Few studies have considered the neural basis of dreaming. We used fMRI, which provides high resolution maps of brain activity, to investigate if functional correlations with the posterior cingulate cortex (PCC) were related to level of detail remembered in a dream. The PCC is hypothesized to be related to dreaming as it is a core hub of the default mode network, a collection of brain regions active during internal mentation (Andrews-Hanna et al. 2010). The data for this exploratory analysis came from an all-night EEG-fMRI study where 12 subjects (8 female) slept two nights in an MRI scanner. At pseudorandom times throughout the night, we delivered increasingly intense auditory tones until participants stated that they were awake. Then, they were asked to describe everything that was going through their mind just before the sound. There were 75 reports from night 2 across three categories: full dreams (mental activity during sleep; n=41), white dreams (cannot remember content; n=17), and no dreams (no reported mental activity during sleep; n=17). Reports were transcribed using Sonix, a web-based software that uses automatic speech recognition and natural language processing, and manually checked. The mean number of words in each report was 19.04 ± 18.48. The skewness of the word count distribution was 1.62 (skewed right), and its kurtosis was 2.43 (platykurtic). The fMRI data were preprocessed using a custom version of the "afni_proc" pipeline from the AFNI software (Taylor et al. 2018). While EEG data were collected, the data were not analyzed in order to explore dreaming correlates beyond the NREM-REM dichotomy (Nir & Tononi 2010). We correlated the signal of the PCC and the rest of the brain in the four minutes before each arousal. We took the absolute value of the correlation and applied a Fisher's Z transformation, then computed a second correlation between the first correlation and word counts using linear mixed effects. The resulting statistical maps were thresholded at a *p*-value of 0.05 (two-sided), and multiple testing correction was applied with a minimum cluster size of 1233 voxels (2.5 x 2.5 x 2 mm resolution) to give a family-wise *p*-value of 0.05. We found one large cluster of 9374 voxels with a center of mass in the cerebellar vermis. The cluster was widespread and included bilateral lingual gyrus, cuneus, and calcarine gyrus (primary visual cortex), as well as hippocampus, thalamus, and posterior cerebellum. Our findings suggest that these visual and memory related areas have stronger functional correlations with the PCC as word count increases. This analysis provides novel evidence for sleep stage independent functional correlations of brain activity with dreaming.

Disclosures: H. Halivni: None. **H. Gura:** None. **J.A. de Zwart:** None. **H. Mandelkow:** None. **F.N. Yang:** None. **P. van Gelderen:** None. **J.H. Duyn:** None. **D. Picchioni:** None.

Poster

PSTR161. Sleep: Relationship to Disease, Disorders, Cognition, and Behavior

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR161.12/JJ1

Topic: F.07. Biological Rhythms and Sleep

Support: New Faculty Start-Up Funds 13-2720-00000-N000844
Tier II Canada Research Chair in Translational Neuroscience and Dementia CRC-2020-00047

Title: Sleep and performance on tests of pattern separation and the Cambridge Neuropsychological Test Automated Battery (CANTAB)

Authors: *A. ROENNINGEN, D. GILL, B. A. KENT;
Dept. of Psychology, Simon Fraser Univ., Burnaby, BC, Canada

Abstract: Sleep disturbances are considered both a dementia risk factor and symptom. The present study aimed to identify cognitive tests sensitive to sleep-dependent cognition. Experiment 1 was a correlational exploratory study and recruited young (N=78; aged 18-30 years) and middle-aged and older (N=39; aged 50 - 100 years) adults. We monitored their sleep patterns over 7 consecutive days using Condor ActTrust 2 actigraphy watches and daily sleep diaries. On day 7, participants completed cognitive testing in the laboratory. The cognitive tests used were the Prodromal Alzheimer's and MCI battery from the Cambridge Neuropsychological Test Automated Battery (CANTAB), as well as the Mnemonic Similarity Task (MST), which taxes pattern separation. The Psychomotor Vigilance Task (PVT) was used as a positive control. Additionally, we assessed the middle-aged and older adults' cognitive performance using the Montreal Cognitive Assessment (MoCA). Experiment 2 examined how one night of total sleep deprivation (TSD) affected cognitive performance using the same cognitive measures. We observed a sleep deprived group (N=16; aged 18-40) overnight in the laboratory. To reduce phase-shifting effects of nighttime light exposure, participants wore UVEX S1933X blue-wavelength blocking glasses. The rested control group (N=28; aged 18-40) slept normally in their home environment. In experiment 1, there were weak positive relationships between total sleep time and MST performance in both the younger and older adults. In experiment 2, sleep deprived participants showed poorer performance on MST measures than rested participants. There was no relationship between CANTAB's pattern separation measure and sleep in the younger or older participants. Data analysis is still ongoing, but these preliminary analyses suggest that performance on MST is affected by sleep. Cognitive tests sensitive to sleep-dependent cognition can be used as clinical trial outcome measures for sleep-promoting treatments.

Disclosures: A. Roenningen: None. **D. Gill:** None. **B.A. Kent:** None.

Poster

PSTR161. Sleep: Relationship to Disease, Disorders, Cognition, and Behavior

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR161.13/JJ2

Topic: F.07. Biological Rhythms and Sleep

Title: Polysomnography and factors associated with daytime sleepiness in older adults with Obstructive Sleep Apnea Syndrome.

Authors: *M. MENDOZA-MELÉNDEZ[1], A. JIMÉNEZ-ANGUIANO[2], U. JIMÉNEZ-CORREA[3], I. VALERIO-ROBLES[3], A. B. E. MENDOZA-BELTRAN[3];
[1]NEUROSCIENCE, UNAM & UAMI, Ciudad de México, Mexico; [2]BIOLOGIA DE LA REPRODUCCION AREA DE NEUROCIENCIAS, UAM I, Av. San Rafael Atlixco 186, Mexico; [3]UNAM, UNAM MEXICO CITY, Mexico

Abstract: Objective: Sleepiness may be considered a symptom or risk factor associated with sleep disorders and physical or mental illness, and is frequently evaluated using the Epworth Sleepiness Scale (ESS). The aim of this study was to determine the polysomnographic changes, prevalence and factors associated with daytime sleepiness in a sample of older adults. **Methods:** A Retrospective Study following the methodology of Medical Record Review of patients 60 years and older at the Sleep Disorder Clinic of the Mexico General Hospital. The measures of association used were chi-squared (χ^2), odds ratios (OR), and a multivariate logistic regression and polysomnographic. **Results:** We found that the prevalence of sleepiness in older adults is very high: three times that of the general population in Mexico. The ESS showed that 57.6% have sleepiness, with a mean score of 13.62±4.44 (to moderate sleepiness). The 81.93% of the patients were diagnosed with Obstructive Sleep Apnea Syndrome (OSAS) which is the most prevalent of sleep disorder in the elderly population. The prevalence of patients with sleepiness diagnosed with OSAS was 50.28% (1.69% mild, 7.34% moderate and 41.24% severed). The risk factors associated with sleepiness in older adults are OSAS diagnosis (OR=3.345), nocturnal snoring interrupted by silence (OR=3.412), daytime naps (OR=3.43), nocturnal sleep disordered breathing (OR=2.185), daytime sleepiness perception (OR=2.756), and use of alcohol to fall asleep (OR=1.805). Our results show a strong association between sleepiness and OSAS. Regression model: (P;; [Y;;=S;;] = 0.80). The older adults with OSAS presented: Total sleep time (363.55 + 76.85, p=0.02), REM sleep duration time (60.91 + 30.93, p= 0.002), lights-off to N2 latency (17.92 + 12.41, p=0.02), sleep efficiency total (75.37 + 15.80, p=0.03), and awakening after sleep initiation (12.64 + 11.69, p=0.03) were significantly higher in AMs with daytime sleepiness. While the latency from lights off to N1 (26.16 + 28.55, p=0.02), to REM (134.11 + 93.93, p=0.005) and awakening before sleep (15.32 + 17.88, p=0.0002) were significantly lower. **Conclusion:** The prevalence of daytime sleepiness in older adults is very high: three times that of the general population in Mexico. OSAS is the most prevalent of sleep disorder in the older adult population in this Sleep Disorder Clinic. The risk factors associated with sleepiness are diagnosis of OSAS, daytime naps, and daytime sleepiness perception. Our results show a strong association between sleepiness and severe OSAS.

Disclosures: M. Mendoza-Meléndez: None. **A. Jiménez-anguiano:** None. **U. Jiménez-Correa:** None. **I. Valerio-Robles:** None. **A.B.E. Mendoza-Beltran:** None.

Poster

PSTR161. Sleep: Relationship to Disease, Disorders, Cognition, and Behavior

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR161.14/JJ3

Topic: F.07. Biological Rhythms and Sleep

Title: Samelisant (SUVN-G3031), a potent and selective histamine 3 receptor inverse agonist for the potential treatment of excessive daytime sleepiness in patients with narcolepsy with and without cataplexy

Authors: *V. BENADE, J. RAVULA, P. JAYARAJAN, V. GOYAL, S. JETTA, A. SHINDE, R. NIROGI, V. JASTI;
Suven Life Sci. Ltd, Hyderabad, India

Abstract: Samelisant (SUVN-G3031) is a potent and selective histamine 3 receptor inverse agonist. In orexin knockout mice, samelisant produced wake-promoting and anticataplectic effects suggesting its potential therapeutic utility in the treatment of narcolepsy. Safety and tolerability studies in animals and healthy human volunteers suggested a favourable risk/benefit profile for samelisant. Samelisant is currently being evaluated as a monotherapy in a double blind randomized controlled trial (Phase-2 proof of concept study) in USA and Canada for treatment of excessive daytime sleepiness in narcolepsy patients with or without cataplexy (ClinicalTrials.gov Identifier: NCT04072380). Subjects diagnosed with narcolepsy as per ICSD-3 criteria, aged between 18 to 65 years with an Epworth Sleepiness Scale (ESS) score of ≥12 and mean Maintenance of Wakefulness Test (MWT) time of <12 min are considered eligible for the study. A total of 171 subjects are to be randomized into one of three treatment arms (placebo, samelisant 2 mg and samelisant 4 mg) in 1:1:1 ratio. Each subject will receive either placebo or samelisant once daily for 2 weeks. The primary efficacy endpoint is change in MWT score from baseline to week 2. Secondary endpoints are change in ESS and CGI-S from baseline to week 2. Safety is being monitored throughout the study by medical monitor and by data safety monitoring committee. The study has completed the recruitment of patients in May-2023. Since the study is not complete, a breakdown of demographics and baseline characteristics by treatment group will not be available until after completion of the study. The median age of subjects is 32 years (range: 18-58 years) with mean BMI of 28.8 kg/m^2 (range: 18.3- 43.9 kg/m2). Overall, 53% subjects were of narcolepsy type-1, 71% were female and 68% were Caucasian. Mean (SD) baseline values of MWT and ESS scores are 6.0 (4.3) and 17.23 (2.8), respectively. Baseline characteristics are consistent with the general narcolepsy population. The data readout from this study is expected in August 2023.

Disclosures: **V. Benade:** A. Employment/Salary (full or part-time); Suven Life Sciences Ltd. **J. Ravula:** A. Employment/Salary (full or part-time); Suven Life Sciences Ltd. **P. Jayarajan:** A. Employment/Salary (full or part-time); Suven Life Sciences Ltd. **V. Goyal:** A. Employment/Salary (full or part-time); Suven Life Sciences Ltd. **S. Jetta:** A. Employment/Salary (full or part-time); Suven Life Sciences Ltd. **A. Shinde:** A. Employment/Salary (full or part-time); Suven Life Sciences Ltd. **R. Nirogi:** A. Employment/Salary (full or part-time); Suven Life Sciences Ltd. **V. Jasti:** A. Employment/Salary (full or part-time); Suven Life Sciences Ltd.

Poster

PSTR161. Sleep: Relationship to Disease, Disorders, Cognition, and Behavior

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR161.15/JJ4

Topic: F.07. Biological Rhythms and Sleep

Support: This study was supported by Cannvalate and the Barbara Dicker Brain Sciences Foundation. The sponsors provided funding for the study only. The study treatments were provided by Brains Bioceuticals.

Title: The effect of cannabidiol on primary insomnia

Authors: *L. A. DOWNEY, A. NARAYAN, A. C. HAYLEY;
Swinburne Univ., Melbourne, Australia

Abstract: Study Objectives: Low dose cannabidiol (CBD) has become readily available in numerous countries; however, little consensus exists on its efficacy as a sleep aid. This trial explored the efficacy of 150mg of CBD (n=15) compared to placebo (n=15) as a sleep aid in primary insomnia. CBD supplementation was hypothesized to decrease insomnia symptoms and improve aspects of psychological health, relative to placebo.Methods: Using a randomized, placebo-controlled parallel design featuring a single-blind placebo run-in week followed by a two-week double-blind dosing phase, participants consumed the assigned treatment sublingually 60-minutes before bed nightly. Wrist-actigraphy and sleep diaries measured daily sleep. Sleep quality, sleep effort and well-being were measured weekly over four in-lab visits with insomnia severity and trait anxiety measured at screening and study conclusion.Results: Insomnia severity, subjective sleep onset latency, sleep efficiency and wake after sleep onset (WASO) did not differ between treatment throughout the trial (all $p>0.05$). Compared to placebo, CBD treatment produced improved well-being throughout the trial (V4 mean difference=2.60, SE 1.20), transient improvements to behavior following wakening (V3 mean difference=3.93, SE 1.53) and superior objective sleep efficiency at V4 (mean difference=6.85, SE 2.95) (all $p<0.05$). At V2, CBD reported less objective WASO but this was not maintained at V4 ($p>0.05$). No other significant treatment effects were observed.Conclusions: Nightly supplementation of 150mg CBD was similar to placebo for the majority of outcomes and effectively improved well-being, suggesting more prominent psychological effects. Additional controlled trials examining longer treatment periods and varying doses are crucial.

Disclosures: L.A. Downey: B. Contracted Research/Research Grant (principal investigator for a drug study, collaborator or consultant and pending and current grants). If you are a PI for a drug study, report that research relationship even if those funds come to an institution.; This study was supported by Cannvalate and the Barbara Dicker Brain Sciences Foundation. The sponsors provided funding for the study only. The study treatments were provided by Brains Bioceuticals.. **A. Narayan:** None. **A.C. Hayley:** None.

Poster

PSTR161. Sleep: Relationship to Disease, Disorders, Cognition, and Behavior

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR161.16/JJ5

Topic: F.07. Biological Rhythms and Sleep

Title: Capturing the effect of habitual sleep characteristics on white matter microstructural properties.

Authors: *H. A. WINDMILL[1], D. GRAHAM[2], A. DHANDA[3], A. D. SMITH[1], M. E. ROSER[4], S. D. HALL[2];
[1]Sch. of Psychology, [3]Peninsula Med. Sch., [2]Univ. of Plymouth, Plymouth, United Kingdom; [4]Sch. of Psychology, Plymouth Univ., Plymouth, United Kingdom

Abstract: Introduction: Many neurological disorders present high levels of fatigue alongside disrupted sleep yet our understanding of the neurological effects of habitual sleep characteristics is poor. Neuroimaging research often focuses on the impact of sleep disorders or deprivation on brain function and structure, commonly evidencing altered activation and microstructural changes. This study aims to investigate effects of habitual sleep by using automated tractography to investigate whether our typical sleep habits and hygiene impair microstructural connectivity in cognitive and motor tracts. **Methods:** Participants (n=30) wore an actigraphy wristwatch (GENEActiv) for 7 days to obtain objective measures of sleep. An MRI protocol consisting of a T1 anatomical scan and diffusion weighted imaging sequence was then acquired. Data was pre-processed, and DTI (FA, AD, RD and ADC) and NODDI (ICVF and ODI) metric maps were created. Key tracts relating to cognitive and motor functioning were selected for automated tractography and average values of each metric were extracted from each tract. A principal component analysis was carried out on objective sleep variables to identify areas of shared variance. 3 principal components (PCs) were identified accounting for 94.8% of overall variance: PC1 (37.3%) informing on sleep chronotype, PC2 (34.1%) informing on sleep efficiency and PC3 (23.4%) informing on rise times. These PCs were then used in Bayesian linear regression models as predictors of microstructural properties. **Results:** Bayesian analysis indicated anecdotal evidence for an effect of sleep chronotype and rise times on both intracellular and extracellular properties of the arcuate fasciculus, corpus callosum, fornix and superior longitudinal fasciculus. Additionally, this study provides moderate evidence for an effect of sleep chronotype on water diffusivity in the corticospinal tract. **Conclusion:** Evidencing the role of habitual sleep characteristics on both axonal and free water properties of cognitive and motor tracts demonstrates the influence of our daily sleeping habits on white matter microstructural properties in a healthy population. Through this understanding, we can further expand on the importance of supporting sleep hygiene when investigating cognitive symptoms of neurological disease.

Disclosures: H.A. Windmill: None. **D. Graham:** None. **A. Dhanda:** None. **A.D. Smith:** None. **M.E. Roser:** None. **S.D. Hall:** None.

Poster

PSTR161. Sleep: Relationship to Disease, Disorders, Cognition, and Behavior

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR161.17/JJ6

Topic: F.07. Biological Rhythms and Sleep

Support: Internal funds and the Gill Endowed Professorship to SS

Title: Effects of thermal neutral exposure on sleep architecture in mice: a closed loop thermoregulatory approach

Authors: *D. IRADUKUNDA, J. WANG, S. SUNDERAM;
F. Joseph Halcomb III, MD, Dept. of Biomed. Engin., Univ. of Kentucky, Lexington, KY

Abstract: Sleep is essential for physiological health and well-being. Temperature is one of many factors that play a significant role in modulating sleep. Both humans and animals exhibit changes in sleep composition and architecture in response to alterations in ambient temperature. In previous work, we have investigated the effect of both static and dynamic changes in ambient temperature on sleep in mice. However, these protocols were implemented mostly without taking the state of vigilance into account. Here, we investigate the effect of thermoneutral temperatures exposure on mouse sleep only after sleep onset. All animal procedures received prior approval from the University of Kentucky IACUC. Male BALB/c wild-type mice (n=4), aged 7-8 months, were selected for the study. Cage temperature was manipulated using a custom-built thermostatic control system consisting of infrared heating lamps that were switched on or off in response to the error in cage temperature with respect to a setpoint. Sleep-wake state was monitored using a piezoelectric ("piezo") motion sensor (Signal Solutions, LLC) placed on the floor of the mouse cage. Sleep was detected when the mean-squared power in the piezo signal in a moving 1-second window dropped below a preset threshold. After allowing the mice to acclimate a 24-hour baseline recording was performed. For the next two days, during the 12-hour light period, the setpoint temperature was elevated to 30℃ (thermoneutral for mice) when the proportion of sleep in a moving one-minute window exceeded 90% and reset to the room temperature of 22℃ when it dropped below 10%. The number of sleep bouts, as well as the percentage time spent in sleep were calculated as outcome measures and compared for the experimental condition against the baseline. Our preliminary results show an increase in percent time spent in sleep and a reduction in sleep fragmentation (greater number of transitions into and out of sleep) for the experimental days compared to the baseline. Specifically, mean sleep time went from 56±12% in the baseline to 58±10%) on treatment Day 1 and 63±13% on treatment Day 2. However, the effects of similar changes in temperature without regard to vigilance state need to be considered. Furthermore, these results are based on sleep-wake discrimination using a motion sensor. Electroencephalographic recordings are planned and will provide further insights into the effect of this protocol on sleep efficiency and sub-states of sleep. This study tests the feasibility of modulating sleep architecture in mice using a closed-loop thermoregulatory approach in preclinical animal models with potential implications for the treatment of sleep-related disorders.

Disclosures: D. Iradukunda: None. **J. Wang:** None. **S. Sunderam:** None.

Poster

PSTR161. Sleep: Relationship to Disease, Disorders, Cognition, and Behavior

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR161.18/JJ7

Topic: F.07. Biological Rhythms and Sleep

Support: American Academy of Sleep Medicine (AASM) Foundation Bridge to Success B2-23-04
Pilot Project from Carolina Autism and Neurodevelopment Research Center

Title: Pregnancy significantly alters the effects of chronic kynurenine on sleep and arousal: Implications for maternal and fetal health outcomes

Authors: *C. J. WRIGHT[1,3], G. L. LOFTUS[3], M. V. PIROLI[3], H. VALAFAR[2], A. POCIVAVSEK[3];
[1]Pharmacology, Physiology, and Neurosci., [2]Computer Sci. and Engin., Univ. of South Carolina, Columbia, SC; [3]Pharmacology, Physiology, and Neurosci., Univ. of South Carolina Sch. of Med., Columbia, SC

Abstract: Pregnant individuals often experience stressors and sleep disturbances that can adversely impact prenatal development and fetal health outcomes. Kynurenic acid (KYNA), an endogenous antagonist of NMDA and α7nACh receptors, is elevated by prenatal insults such as sleep deprivation. In adult rodents, transient KYNA elevations disrupt sleep-wake behavior. As KYNA levels are also elevated in the brain of patients with neurodevelopmental disorders (NDDs) including schizophrenia and bipolar disorder, we presently sought to determine how prenatal KYNA elevations impact maternal sleep. We induced a prenatal insult, the embryonic kynurenine model, by feeding pregnant rats (N=6) chow laced with the direct KYNA bio-precursor kynurenine (100 mg/day) to stimulate prenatal KYNA elevation from embryonic day (ED) 15 to ED 22. Control dams (N=6) were fed unlaced chow, while separate non-pregnant females (N=5) received a kynurenine-laced diet for 8 days. Sleep was recorded during the treatment with radio telemetry devices to acquire EEG/EMG. Before treatment or pregnancy, each animal underwent four days of baseline recordings. Vigilance states were classified as wake, rapid eye movement (REM) sleep, and non-REM (NREM) sleep. Our findings revealed that chronic kynurenine treatment in non-pregnant females significantly reduced light phase REM sleep duration ($P<0.05$) and increased dark phase NREM sleep duration ($P<0.05$). In pregnant rats, irrespective of kynurenine treatment, we observed significantly increased total sleep duration in the dark phase ($P<0.05$) until ED 20, at which point kynurenine-treated mothers experienced significantly reduced dark phase total sleep duration compared to pregnant controls ($P<0.05$) and reduced REM theta power (frequency x pregnancy interaction, $P<0.0001$). We also observed reduced dark phase relative cage activity in pregnant controls but not pregnant or non-pregnant kynurenine-treated females. Taken together, we observed that pregnancy significantly alters the effects of chronic kynurenine treatment on sleep-wake behaviors in rats. Our data indicate that kynurenine treatment during pregnancy impedes restorative sleep and supports our hypothesis that elevations in KYNA affect sleep. Future studies will evaluate how pregnancy and

kynurenine treatment affect KYNA levels in maternal saliva, plasma, and brain as well as the influence of sex on fetal tissue kynurenine pathway metabolism. Our novel findings serve to better our understanding of the etiology of NDDs and inform future work on a novel potential therapeutic target, to reduce KYNA, for preventative treatments in individuals suffering from sleep disruptions.

Disclosures: C.J. Wright: None. **G.L. Loftus:** None. **M.V. Piroli:** None. **H. Valafar:** None. **A. Pocivavsek:** None.

Poster

PSTR161. Sleep: Relationship to Disease, Disorders, Cognition, and Behavior

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR161.19/JJ8

Topic: F.07. Biological Rhythms and Sleep

Support: Veterans Administration (Merit award#IBX00261)
National Institute of Health (#AA028175)
MU Research Council(URC-22-006)

Title: Effects of antisense-induced downregulation of circadian gene Period1 in the basolateral amygdala on spontaneous sleep-wakefulness and consolidation of traumatic memory

Authors: *M. PARIKH[1], R. SHARMA[2], A. CHISCHOLM[1], P. SAHOTA, 65201[3], M. THAKKAR[4];
[1]Univ. of Missouri, Columbia, Columbia, MO; [2]Univ. of Missouri, Columbia, MO; [3]Univ. of Missouri; Harry S Truman veteran affairs Hosp., Columbia, MO; [4]HSTMV Hospital/University of Missouri, Columbia, MO

Abstract: Effects of antisense-induced downregulation of circadian gene *Period 1* in the basolateral amygdala on spontaneous sleep-wakefulness and consolidation of traumatic memory BACKGROUND The basolateral amygdala (BLA) is implicated in the regulation of emotional processes. Recently it has been shown that the BLA has a role in sleep-wakefulness. Considering the significance of the BLA in sleep-wakefulness and traumatic memories, we asked! Does the circadian gene Period 1 (Per1) within the BLA contributes to sleep-wakefulness and in the consolidation of traumatic memories? Thus, we hypothesize that downregulation of Per1 in the BLA will result in changes in sleep wakefulness and impaired traumatic memory consolidation. **METHODS** Adult male C57BL/6J mice were stereotaxically implanted with three screw electrodes on the skull for recording EEG and three wire electrodes in nuchal muscle to record muscle activities (EMG). In addition, three Teflon-coated tungsten wire electrodes (76 µm dia) along with bilateral stainless-steel guide cannulas were also implanted (2.0) mm above the BLA region. The animals were connected to the recording setup and allow to habituate for 7-day period. ***Experiment 1:*** The mice were divided into two groups: Antisense and Saline. Antisense (experimental group) and saline solutions (control group) were respectively infused

through the guide cannulas into the BLA on light-onset. Spontaneous sleep-wakefulness was recorded for following 48 hours. ***Experiment 2:*** One hour after light-onset, contextual training was performed with contextual cage as conditioned stimulus (CS;5 min) and soiled cat litter as unconditioned stimulus (US;10 min) on Day-1 followed by memory recall testing on day-2. LFP were recorded from the BLA. **RESULTS** ***Experiment 1****:* Knockdown of Per1 in the BLA reversed sleep-wake cycle with increased wake and reduced sleep during the light period, increased sleep, reduced wake during dark period. ***Experiment 2:*** The effect of Per1 downregulation in BLA on contextual training, is ongoing. **CONCLUSION** Initial studies suggest that antisense-induced downregulation of Per1 in the BLA, reverses sleep-wake cycle, has effect on traumatic memory.

Disclosures: M. Parikh: None. **R. Sharma:** None. **A. Chischolm:** None. **P. Sahota:** None. **M. Thakkar:** None.

Poster

PSTR161. Sleep: Relationship to Disease, Disorders, Cognition, and Behavior

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR161.20/JJ9

Topic: F.07. Biological Rhythms and Sleep

Support: NIH grant AG061824.

Title: Sleep and explicit memory impairments and blood-brain barrier permeability induced in naïve mice by fecal microbiota transplantation from mice exposed to chronic sleep fragmentation.

Authors: *C. PUECH, M. B. BARROW, M. BADRAN, D. GOZAL;
Child Hlth. Res. Inst., Univ. of Missouri, Columbia, MO

Abstract: Obstructive sleep apnea (OSA) is a chronic and highly prevalent condition characterized by intermittent hypoxia (IH) and sleep fragmentation (SF) and is associated with a large spectrum of morbidities including excessive sleep propensity (EDS) and cognitive deficits. SF-induced cognitive declines are negatively correlated with increased Blood Brain Barrier (BBB) permeability. Both SF and IH alter the gut microbiome (GM). Furthermore, fecal microbiota transfer (FMT) from mice exposed to chronic IH mimicking moderate-severe OSA into naïve mice recapitulates the sleep disturbances induced by IH. We hypothesized than FMT from SF-exposed mice can also induce sleep perturbations and will result in explicit memory dysfunction via BBB dysfunction.Male C57Bl/6J 8-week-old mice were housed in custom-designed cages with a silent motorized mechanical sweeper traversing the cage floor at 2-min intervals (SF) 12 hours/day for 4 weeks. Fecal samples were collected and frozen. C57Bl/6J naïve mice (male and female) were randomly assigned to a validated FMT protocol by gavage for 6 weeks with either SF or SC fecal slurry. Sleep activity was recorded using a non-invasive, high throughput piezoelectric system, and sleep states were automatically scored by validated

AI-derived computer algorithms. Cognitive function was evaluated using the novel object recognition (NOR) test which provides reliable assessments of explicit declarative memory. BBB permeability was assessed by 4KDA-dextran-FITC iv injection, and FITC brain immunofluorescence was quantified.Dark phase sleep percentage was significantly increased in FMT-SF mice (39± 7 %) when compared to FMT-SC mice (27 ± 11 %, p= 0.014) with no differences in light phase and total sleep. FMT-SF mice exhibited reduced interest for novelty in NOR tests compared to FMT-SC mice (preference scores: FMT-SF: 60 ± 13% vs FMT-RA: 75 ± 20% p =0.0427). BBB permeability in FMT-SF mice was increased (2.660 ±0.2µg of dextran/g brain in comparison to FMT-RA mice (2.0 ±0.5µg of dextran/g brain, p=0.0139).Fecal GM transfer from mice exposed to SF induces sleep disturbances and deficits in explicit memory in naïve recipient mice that recapitulate the effects of SF. Furthermore, these adverse effects seem to be associated at least in part with altered BBB permeability.

Disclosures: C. Puech: None. **M.B. Barrow:** None. **M. Badran:** None. **D. Gozal:** None.

Poster

PSTR161. Sleep: Relationship to Disease, Disorders, Cognition, and Behavior

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR161.21/JJ10

Topic: F.07. Biological Rhythms and Sleep

Support: NIH Grant DA042110

Title: Insomnia treatment drug lemborexant rescues REM sleep dysfunction associated with methamphetamine vapor withdrawal

Authors: *G. P. HUFFCUTT, M. R. JONES, B. E. SCHMEICHEL;
Biomed. Sci., East Tennessee State Univ., Johnson City, TN

Abstract: In 2020, 2.5 million people abused methamphetamine (MA) in the US. Chronic MA use can lead to disordered sleep, particularly during withdrawal, and clinical studies show that sleep dysfunction is a strong predictor for relapse. The neuropeptide hypocretin (HCRT) plays a critical role in the transition to wakefulness and also modulates drug reward. Enhanced HCRT signaling in the brain underlies the sleep disorder insomnia and the dual-HCRT-receptor antagonist lemborexant (LEM) is prescribed for treatment of insomnia in humans. Here we characterized sleep dysfunction associated with MA vapor (MAV) withdrawal in rats and hypothesized that HCRT signaling contributes to poor sleep. Adult male Wistar rats (N =8) received a telemetry device implant and EEG/EMG signals were recorded for 23 hours (12h light:11 h dark). Rats were exposed to passive MAV for 4 weeks to induce dependence. Rats showed significant decreases in body temperature and novel object recognition during acute drug withdrawal (1-day post MAV). Sleep/wake data were analyzed prior to MAV exposure (baseline; BL), during withdrawal (1 week of MAV abstinence), and during protracted abstinence (four weeks of MAV abstinence). LEM (0 and 30 mg/kg, counter-balanced) was

administered at the beginning of the light cycle prior to sleep recordings during abstinence. After 1 week of abstinence, rats showed a decrease in rapid eye movement (REM) sleep time in the light cycle during withdrawal, and there was an increase in REM sleep time during the dark cycle, indicating possible REM sleep rebound. The number of bouts of REM sleep decreased during the light cycle with no change in average bout duration. LEM restored the amount of REM sleep time and the number of REM sleep bouts during the light cycle. Although the total amount of non-REM (NREM) time was unchanged, average NREM bout duration decreased and number of bouts increased in the dark cycle during withdrawal, indicating NREM sleep was more fragmented during the dark cycle. LEM had no significant effect on NREM sleep. Rats also showed no change in total amount of wakefulness (WAKE) during abstinence, but did show an increase in the number of WAKE bouts during the light and dark cycles. After 4 weeks of abstinence, REM sleep time remained reduced, trending toward significance ($p=0.08$); number of REM bouts in the light cycle and REM sleep time in the dark cycle returned to BL levels. While the mean NREM bout duration remained reduced, there were no changes in WAKE measures during protracted abstinence compared to BL. Overall, these findings show that REM and NREM sleep are dysregulated during abstinence from MAV and that HCRT neurotransmission contributes to the disrupted sleep.

Disclosures: G.P. Huffcutt: None. **M.R. Jones:** None. **B.E. Schmeichel:** None.

Poster

PSTR161. Sleep: Relationship to Disease, Disorders, Cognition, and Behavior

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR161.22/JJ11

Topic: C.07. Neurotoxicity, Inflammation, and Neuroprotection

Support: NIH AG054104
NIH AG064231

Title: Chronic sleep disruption disturbs behavioral state-dependent calcium dynamics in hippocampal CA1 CAMKII neurons

Authors: *A.-H. CHOU, P. FENIK, H. HOLLIS, C. LY, B. T. KEENAN, R. C. ANAFI, S. C. VEASEY;
Univ. of Pennsylvania, Philadelphia, PA

Abstract: Chronic sleep disruption (CSD) in young adult wild type mice leads to irreversible injury to and dysfunction of hippocampal CA1 CAMKII neurons. Yet mechanisms underlying these injuries are not known. Sleep/wake states influence calcium (Ca^{2+}) dynamics in CA1 CAMKII neurons, which could then influence neuronal survival. We predicted that chronic sleep fragmentation (CSF), a form of CSD, would perturb behavioral state-dependent Ca^{2+} dynamics in CAMKII neurons of the CA1 region. Humanized amyloid precursor protein knock-in mice (hAPP) (n= 5F Rested, n= 4F CSF) received AAV-CAMKII-GCaMP6f injections in the CA1, as

well as electroencephalography, electromyography, and GRIN lens implantations at 6-9 months of age. Mice were randomized to CSF or control sleep conditions for 16 weeks. We then conducted baseline *in vivo* Ca^{2+} imaging across three behavioral states: quiet wake, exploratory wake, and non-rapid eye movement (NREM) sleep. We developed and validated a Shiny R application to robustly capture Ca^{2+} transient amplitude and duration criteria for a transient. We analyzed the relative fluorescence per neuron across time ($\Delta F/F$) and Ca^{2+} transient frequency between rested and CSF mice. We compared differences in overall and sleep-stage-specific calcium dynamics between mice exposed to CSF or rested animals using linear mixed effects models with a random mouse effect to account for repeated measurements (e.g., cells) per animal; similar models were used to compare overall differences between sleep stages. Results revealed no significant differences between CSF and rested mice across combined behavioral states for $\Delta F/F$ ($p=.65$) and frequency ($p=.70$). However, CSF significantly increased $\Delta F/F$ in NREM sleep ($p<.05$) and quiet wake ($p<.05$) periods, and decreased $\Delta F/F$ in exploratory wake ($p<.0001$). Further, frequencies increased during sleep ($p<.001$). We next examined sleep/wake characteristics in a separate cohort of mice after rested and CSF exposures. Although CSF did not affect % time in Wake or REM sleep, we did see a reduction in % time in NREM sleep. However, there were no differences in arousal frequencies or in the duration or numbers of NREM sleep bouts, suggesting there are no major differences in NREM sleep consolidation after CSF. Together, our results show that CSF disrupts behavioral state influences on Ca^{2+} dynamics in CAMKII CA1 neurons and that these effects are independent of effects of CSD on behavioral state. Both an increase in activity across NREM sleep and reduced activity in active exploratory wakefulness may contribute to CSD-induced hippocampal dependent memory impairments.

Disclosures: **A. Chou:** None. **P. Fenik:** None. **H. Hollis:** None. **C. Ly:** None. **B.T. Keenan:** None. **R.C. Anafi:** None. **S.C. Veasey:** None.

Poster

PSTR161. Sleep: Relationship to Disease, Disorders, Cognition, and Behavior

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR161.23/JJ12

Topic: C.10. Brain Injury and Trauma

Support: NIHAG054104
NIHAG064231

Title: Extended wakefulness alters calcium transient dynamics and morphology in hippocampal CA1 CaMKII neurons

Authors: *C. LY[1], P. FENIK[1], H. HOLLIS[1], A.-H. CHOU[1], B. T. KEENAN[2], R. C. ANAFI[1], S. C. VEASEY[1];
[1]Ctr. for Sleep and Circadian Neurobio., [2]Biostatistics Core, Univ. of Pennsylvania, Philadelphia, PA

Abstract: Chronic sleep disruption imparts lasting CA1 CaMKII neuron loss and hippocampal memory impairments. The mechanisms by which the injury occurs are not known. CA1 Ca2+ transients show state-behavioral dependency, with higher firing rates in active wake. Two common forms of sleep disruption are chronic sleep fragmentation (CSF), as observed in various sleep disorders including sleep apnea; and short sleep with prolonged periods of active wake (SS), as seen in shift workers. We have found in preliminary reports that CSF influences behavioral state-dependent CA1 CaMKII Ca2+ dynamics. We hypothesized that a period of SS would further disturb CA1 CaMKII neuron Ca2+ dynamics. Here, we explored potential interactions between these two common forms of sleep disruption. Randomized female adult mice with humanized amyloid precursor protein knock-in were subjected to conditions of control rest (n=5) or CSF (n=4). Mice received a CA1 AAV-CAMKII-GCaMP6F injection and were later implanted with a CA1 GRIN lens and electroencephalographic (EEG) and electromyographic (EMG) electrodes to ascertain behavioral state in parallel with Ca2+ dynamics. We examined Ca2+ transient characteristics (ΔF/F, transient frequency, accumulation/clearance duration) across three activity states (wake, active wake, and NREM sleep) at baseline conditions and again after three consecutive days of 8-hour active wake across the lights-on period (SS) in rested and CSF mice. After SS, CSF mice showed increased ΔF/F in sleep ($p<.0001$) and in wake ($p<.05$), but decreased ΔF/F in active wake ($p<0.0001$) compared to rested conditions. These findings, following exposure to SS after rested and CSF conditions, showed the same directionality as observed responses prior to SS; however, increases in wake and sleep ΔF/F were more robust, and differences in active wake were less pronounced. A significant reduction in the Ca2+ transient frequency in active wake was only observed in CSF mice after SS ($p<.001$). In contrast, prior to SS, active wake transient frequency between CSF and rested conditions was unchanged. Notably, Ca2+ transient architecture changed only in response to SS. Specifically, after SS, we observed prolongation of the clearance duration ($p<.05$) in sleep, and we noted a significant increase in the average spike area ($p<.05$) in quiet wake. Neither effect on Ca2+ transient morphology was observed prior to SS. These findings suggest that these two common forms of sleep disruption, SS and CSF, influence Ca2+ dynamics differentially, and thereby underlying neural mechanisms are expected to be distinct.

Disclosures: C. Ly: None. **P. Fenik:** None. **H. Hollis:** None. **A. Chou:** None. **B.T. Keenan:** None. **R.C. Anafi:** None. **S.C. Veasey:** None.

Poster

PSTR161. Sleep: Relationship to Disease, Disorders, Cognition, and Behavior

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR161.24/JJ13

Topic: C.10. Brain Injury and Trauma

Support: DoD, 308430 USUHS
DoD, APG-70-12224 USUHS
CHIRP-70-3925, USU/NHLBI

Title: Sleep, neurovasculature and microglia at high altitude

Authors: V. TSYTSAREV[1,2], K. WHITING[1,2], M. WANG[3], F. W. LISCHKA[1,2], T. BARVIR[1,2], X. XU[1,2], S. JAISWAL[1,2], N. P. CRAMER[1,2], C. BROWNE[1,2], D. L. DICKSTEIN[1,2], D. P. PERL[2], G. YU[3], ***Z. GALDZICKI**[4];
[1]Sch. of Medicine, Uniformed Services Univ., Bethesda, MD; [2]The Henry M. Jackson Fndn. for the Advancement of Military Hlth. Inc., Bethesda, MD; [3]Virginia Tech., Arlington, VA; [4]Dept. of Anat, Physiol & Genetics, Sch. of Medicine, USU, Bethesda, MD

Abstract: Hypobaric hypoxia caused by high altitude (HA) can lead to impaired tissue oxygenation causing cardiovascular & metabolic dysfunction and cognitive deficits, often with long-lasting effects that persist even after return to normoxic conditions. Previously we have reported that chronic (12 week) HA exposure results in enhanced angiogenic activity, disruption of the blood brain barrier, increased microglia phagocytosis, as well as functional deficits in hippocampal dependent memory tasks (Cramer et al. 2019). These changes can significantly impact sleep. Here, sleep patterns and vascular changes in mice exposed to HA were evaluated. Briefly male C57BL/6J mice were placed in a hypobaric chamber at a simulated altitude of 5000 m (10-11% O_2, ~60% SpO_2) for 3-weeks. In order to assess sleep, polysomnographic recordings were carried out using an EEG/EMG telemetry system (HD-X02 implants, DSI, MN; n=6 per group). Throughout HA exposure, animals exhibited an increase in percent time awake during the light cycle (ZT 12-24), increased REM during the dark cycle (ZT 0-12), and decreased SWS in both phases of the light cycle (2-way ANOVA, $p < 0.001$). Assessment of vasculature was achieved through transcardial perfusion of a high contrast agent, BriteVu (Scarlet Imaging, UT), followed by *ex-vivo* microCT imaging using Bruker Skyscan 1172 (n=4 per group), and analysis using Vesselucida software (MBF, VT) to quantify vascular changes. A main effect of HA was observed on vasculature morphology, with increased total blood volume of vessels in HA brains, likely due to increased average vessel diameter. Tortuosity of blood vessels in HA brains was also increased. Since our previous work demonstrated that HA promoted a neuroinflammatory transcriptome profile and enhanced microglia phagocytic activity, *ex vivo* analysis of microglia dynamics in brain slices were conducted. This study revealed that baseline homeostatic microglia activity (tip number and velocity) was not altered in slices from mice exposed to HA. When the microglia were activated by localized laser ablation of microglia or capillaries, there was a significant attenuation in the degree of ramification of microglia processes in mice exposed to HA. In summary, HA exposure significantly impacts sleep architecture, vasculature density and structure, and alterations in microglia physiology. Future studies should address treatment modalities to alleviate the persistent neurological symptoms associated with HA. Disclaimer: The views expressed here are those of the authors and do not reflect the official policy or position of the U.S. government or the Department of Defense. The authors have no conflict of interest.

Disclosures: V. Tsytsarev: None. **K. Whiting:** None. **M. Wang:** None. **F.W. Lischka:** None. **T. Barvir:** None. **X. Xu:** None. **S. Jaiswal:** None. **N.P. Cramer:** None. **C. Browne:** None. **D.L. Dickstein:** None. **D.P. Perl:** None. **G. Yu:** None. **Z. Galdzicki:** None.

Poster

PSTR162. Mechanisms of Fear and Extinction Memory Modification

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR162.01/JJ14

Topic: G.01. Fear and Aversive Learning and Memory

Support: NSERC Discovery Grant

Title: The contribution of white matter plasticity to promote enrichment-induced forgetting of a contextual fear memory

Authors: *J. J. MAK[1,3], L. M. GAZDZINSKI[1], X. LIN[1], J. G. SLED[2,4], B. J. NIEMAN[2,4], A. L. WHEELER[1,3];
[1]Neurosci. & Mental Hlth., [2]Translational Med., The Hosp. For Sick Children, Toronto, ON, Canada; [3]Dept. of Physiol., [4]Dept. of Med. Biophysics, Univ. of Toronto, Toronto, ON, Canada

Abstract: Forgetting occurs when patterns of neural activity present at memory encoding cannot be reliably reactivated. The precise timing of activity in neural circuits is regulated by white matter properties such as the length and width of axons, as well as the distribution and thickness of myelin that surrounds axons. Living in enriched environments has been shown to promote white matter plasticity, which may disrupt previously established activity patterns associated with specific memories and facilitate forgetting. The objective of this study is to describe the contribution of white matter plasticity to forgetting. The impact of white matter plasticity on contextual fear memory was assessed by housing mice in enriched cages after a single conditioning session where the mice received three 0.75mA foot shocks. Enriched cages consisted of larger, double-decker cages containing both a running wheel and a reconfigurable maze that was rearranged weekly. Food and water were separated at opposite ends of the maze to force mice to constantly relearn how to manoeuvre through each maze arrangement. After living in an enriched environment for four weeks, mice were returned to the conditioning context and freezing was measured as an index of memory. *Ex vivo* mouse brains were scanned on a 7T Agilent MRI system with a diffusion weighted acquisition. Microstructural properties of white matter tracts defined by a segmented atlas were assessed with fractional anisotropy (FA), a measure of white matter integrity derived from diffusion tensor imaging. Effects of enrichment on freezing, white matter FA, and associations between freezing and FA were assessed with linear mixed effects models. Mice housed in enriched environments demonstrated forgetting as measured by reduced time spent freezing during the fear conditioning test compared to mice housed in standard cages (β=-51%, $p=2x10^{-6}$). Enriched housing was associated with altered white matter microstructure, where FA was increased in the corpus callosum, fimbria, and anterior commissure compared to mice housed in standard cages (10% FDR). In mice housed in enriched environments, increased FA was associated with reduced freezing in the corpus callosum and fimbria ($p<0.05$), suggesting that in these regions, white matter plasticity induced by enrichment contributes to forgetting. Altogether, these results support that the plasticity occurring in white matter tracts important for contextual fear memory is contributing to enrichment-induced forgetting.

Disclosures: J.J. Mak: None. **L.M. Gazdzinski:** None. **X. Lin:** None. **J.G. Sled:** None. **B.J. Nieman:** None. **A.L. Wheeler:** None.

Poster

PSTR162. Mechanisms of Fear and Extinction Memory Modification

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR162.02/JJ15

Topic: G.01. Fear and Aversive Learning and Memory

Support: NINDS Grant T32NS105602
NIH Grant 1R01MH122742
Internal funding from UW-Madison School of pharmacy/Medicine and Public Health

Title: Differential modulation of threat assessment by psilocybin and DMT

Authors: *J. RAZIDLO[1], J. NERAD[2], N. JONES[3], C. J. WENTHUR[4];
[1]Neurosci. Training Program, UW-Madison, Madison, WI; [2]Sch. of Pharm., Univ. of Wisconsin-Madison, Madison, WI; [3]Univ. of Wisconsin - Sch. of Pharm., Univ. of Wisconsin, Madison, WI; [4]Univ. of Wisconsin Madison, Univ. of Wisconsin - Madison. Psychoactive Pharmaceut. Investigation Program, Madison, WI

Abstract: Psychiatric disorders such as Major Depressive Disorder (MDD) have become an increasingly prevalent issue and are compounded by the limited efficacy of current treatments. To combat these disorders, researchers have focused on novel therapeutic approaches. Two promising treatments are the serotonergic $5HT_{2A}$ agonists and classical psychedelics, psilocybin and N,N-dimethyltryptamine (DMT). These compounds have been shown to exert rapid antidepressant effects through mechanisms that have yet to be fully understood. These compounds have been observed to induce an acute stress response, as measured by elevated glucocorticoid release after administration. We propose that the therapeutic effects of these drugs are due to a transient enhancement of neural plasticity in the ventral hippocampal medial-prefrontal-cortex (HPC-mPFC) pathway and subsequent memory formation produced by this acute stress. Impaired plasticity in the HPC-mPFC pathway is observed in patients with MDD, and individuals have reported decreased depressive symptoms after taking psychedelics. These improvements in mood have been reported to last weeks to months. Despite this, there are no studies to date that assess the duration of this critical window. To study this window of plasticity at the behavioral level, we used C57BL/6J mice and the associative learning task of fear conditioning. Mice were administered a single intraperitoneal (IP) injection of saline, psilocybin (3 mg/kg), or DMT (10mg/kg) prior to fear conditioning (FC). In both cued and contextual FC there was no difference in freezing behavior in fear acquisition, nor fear extinction between mice that received saline or psilocybin. Interestingly, male mice that received psilocybin showed reduced fear renewal (unpaired t-test, $p=0.0135$), which was not seen in females. What is even more striking is that an impairment of fear extinction was seen in animals that received DMT. Further research is needed to understand any sex dependent effects of psychedelics, as well as dose and timing of drug administration.

Disclosures: **J. Razidlo:** None. **J. Nerad:** None. **N. Jones:** None. **C.J. Wenthur:** B. Contracted Research/Research Grant (principal investigator for a drug study, collaborator or consultant and pending and current grants). If you are a PI for a drug study, report that research relationship even if those funds come to an institution.; Psilera Inc., Mike and Mary Shannon. Other; Usona Institute.

Poster

PSTR162. Mechanisms of Fear and Extinction Memory Modification

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR162.03/JJ16

Topic: G.01. Fear and Aversive Learning and Memory

Support: National Institute on Drug Abuse - psilocybin

Title: Low-dose psilocybin sex-dependently enhances fear extinction in adult rats

Authors: ***C. E. MILLER**, M. M. NAYLOR, C. R. DEL VALLE, H. R. SPARKMAN, B. E. BRAMLAGE, L. P. PUPPEL, M. L. BROWN, A. K. AL-OLIMAT, K. M. KING, M. J. SAVINI, P. R. ZOLADZ;
Psychology, Ohio Northern Univ., Ada, OH

Abstract: Fear-related psychological disorders, such as specific phobias and post-traumatic stress disorder (PTSD), represent a major public health issue. It is thought that these disorders develop, at least in part, through fear conditioning processes. Thus, treatment often involves exposure therapy, a technique based on the concept of extinction. However, this type of therapy is ineffective for many individuals. To augment extinction learning, researchers have paired exposure therapy with pharmacological agents that enhance neuroplasticity (e.g., D-cycloserine), but the results of such manipulations have been inconsistent. It is possible that psychedelics, which have been largely unexplored in their ability to enhance extinction learning, could aid in exposure therapy. Indeed, preclinical research suggests that several psychedelic substances, such as MDMA and DMT, enhance synaptic plasticity. Thus, we explored the dose-dependent effects of the 5-HT_{2A} agonist psilocybin on fear extinction in adult rats. On Day 1, adult male and female Sprague-Dawley rats were habituated to a fear conditioning chamber (Context A). Habituation involved a 3-min acclimation phase, followed by five tone (10-sec, 2 kHZ) presentations [60-sec interstimulus intervals (ISIs)]. On Day 2, the rats underwent fear conditioning in the same context. This involved the same parameters as habituation, except each tone co-terminated with a 1-sec, 1 mA footshock. On Day 3, rats were injected intraperitoneally with psilocybin (0.3 or 1 mg/kg) or vehicle (0.9% saline) 30 min prior to undergoing cue-based fear extinction in a novel environment (Context B). During extinction, the rats were given a 3-min acclimation phase, followed by 30 tone presentations (60-sec ISIs). On Day 4, rats underwent extinction recall by being placed in Context B; the test began with a 3-min acclimation phase, followed by 10 tone presentations (60-sec ISIs). Freezing behavior was quantified by FreezeFrame software (Actimetrics, Inc.). Analyses of freezing behavior during

training and early extinction demonstrated that all rats developed strong fear of the tone. Most importantly, the low dose of psilocybin enhanced extinction learning in males but slowed extinction learning in females. This differential impact of psilocybin on extinction was maintained during extinction recall the next day. Our findings suggest that a low dose of psilocybin augments extinction learning in males, but not females. The sex-dependent nature of this effect warrants additional research.

Disclosures: C.E. Miller: None. **M.M. Naylor:** None. **C.R. Del Valle:** None. **H.R. Sparkman:** None. **B.E. Bramlage:** None. **L.P. Puppel:** None. **M.L. Brown:** None. **A.K. Al-Olimat:** None. **K.M. King:** None. **M.J. Savini:** None. **P.R. Zoladz:** C. Other Research Support (receipt of drugs, supplies, equipment or other in-kind support); National Institute on Drug Abuse.

Poster

PSTR162. Mechanisms of Fear and Extinction Memory Modification

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR162.04/JJ17

Topic: G.01. Fear and Aversive Learning and Memory

Title: Impact of oxytocin pretreatment on signs of fear in the rat

Authors: *D. P. DABERKOW, J. E. ALVAREZ, Jr.;
Biol., Eastern Washington Univ., Cheney, WA

Abstract: Oxytocin has been used as a post-treatment for PTSD. Previous research suggests that pre-treatment of intranasal oxytocin attenuates signs of fear in rats. To further this research, we investigated the specific dose of oxytocin, administered (i.p), that best alleviates signs of fear in rats. Male Sprague-Dawley rats, were split into five experimental groups (n=8 per group): 1.) control group treated with vehicle and no foot shock, 2.) shock group treated with vehicle and received foot shock, 3.) low dose group that received 0.03 mg/kg oxytocin and foot shock, 4.) medium dose group that received 0.3 mg/kg oxytocin and foot shock, and 5.) high dose group that received 1.0 mg/kg oxytocin and foot shock. Rats were treated with oxytocin (or vehicle) 30 min prior to exposure to fear conditioning. On day 1 of fear conditioning, rats were put into a Colbourn Precision fear-conditioning shock chamber. The shock chamber was a plexiglass box (30 cm x 30 cm) with a metal grate floor, which delivered five foot shocks at an intensity of 0.6 mA. Twenty-four hours later, the rats were re-exposed to the chamber for 5 minutes, not shocked, and the freezing time was recorded via a motion detector. Preliminary data suggest that oxytocin pretreatment decreases freezing relative to the untreated shock group. These results suggest that oxytocin, administered i.p., can be used as a prophylactic pre-treatment to mitigate signs of fear.

Disclosures: D.P. Daberkow: None. **J.E. Alvarez:** None.

Poster

PSTR162. Mechanisms of Fear and Extinction Memory Modification

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR162.05/JJ18

Topic: G.01. Fear and Aversive Learning and Memory

Support: Roche Postdoc Fellowship (Project RPF-ID:285
Marie Heim Vögtlin subsidy from Swiss National Science Foundation (PMPDP3 164468)
Swiss National Science Foundation
Synapsis Foundation

Title: Social buffering switches fear to safety encoding by oxytocin recruitment of central amygdala "buffer neurons"

Authors: Y. TANG[1], C. HEGOBURU[1], R. NIU[2], S. GHOSH[2], R. TRIANA DEL RIO[2], I. SALGADO[2], M. ABATIS[2], D. MOTA CASEIRO[2], E. H. VAN DEN BURG[2], C. GRUNDSCHOBER[3], ***R. STOOP**[4];
[1]Contributed equally, Lausanne Univ. Hosp. Ctr., [2]Lausanne Univ. Hosp. Ctr., Ctr. for Psychiatric Neurosci., Prilly, Lausanne, Switzerland; [3]Roche Innovation Ctr., Basel, Switzerland; [4]Univ. of Lausanne, Prilly, Lausanne, Switzerland

Abstract: In behavioral therapy, social support can play an important role in immediate as well as long-term reduction of stress and anxiety. The underlying neural systems remain, however, still largely unknown. Rodent behavioral studies have successfully modeled the fear-reducing effects of social support, referred to as the "Social Buffering of Fear" (SBF). In the present study, we show that signaling of the nonapeptide oxytocin in the central amygdala plays a crucial role in the acute (aSBF) as well as retention of SBF (rSBF). We therefore auditory fear conditioned (FC) rats on Day 1 to a CS1 (5 kHz) and a CS2 (15 kHz) and we tested aSBF by re-exposing them on Day 2 to CS1 either in the absence or presence of a companion and tested rSBF by re-exposing on Day 3 both groups to CS1 and CS2 without the companion. aSBF and rSBF developed specifically to CS1 but not CS2. With pharmacological, chemogenetic, and optogenetic interventions we found that aSBF and rSBF required OT signaling from the paraventricular nucleus of the hypothalamus (PVN) to the central amygdala (CeA). Optogenetic activation of OTergic neurons in the PVN or their projections in the CeA induced aSBF on Day 2 but not rSBF on Day 3. Multi single unit in vivo recordings with optrodes in the PVN and CeA (after infecting PVN OT neurons with ChR2) revealed on Day 2 "fear-encoding" CeA neurons, which increased CS1 responses after FC, and decreased CS1 responses during aSBF as well as rSBF. We also found a second group of CeA neurons that developed responses to CS1 only after aSBF on Day 2 which persisted during rSBF on Day 3. Their activity was excited by blue light stimulation of OTergic PVN projections and by exposure to the companion, similar to PVN OT neurons. Taken together, these findings suggest an inhibitory circuitry in the CeA between OT sensitive "buffer" neurons that inhibit "fear-encoding" neurons during aSBF by OT released from the PVN. aSBF subsequently induces rSBF on Day 3 by switching CS1 responsiveness from "fear-encoding" neurons to "buffer" neurons. OT thus seems required to induce long-lasting

changes in neuronal circuitry in the CeA. Our findings may represent an animal model for studying the circuitry that underlies the long-term reduction of stress and anxiety by social support.

Disclosures: **Y. Tang:** None. **C. Hegoburu:** None. **R. Niu:** None. **S. Ghosh:** None. **R. Triana Del Rio:** None. **I. Salgado:** None. **M. Abatis:** None. **D. Mota Caseiro:** None. **E.H. van den Burg:** None. **C. Grundschober:** None. **R. Stoop:** None.

Poster

PSTR162. Mechanisms of Fear and Extinction Memory Modification

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR162.06/JJ19

Topic: G.01. Fear and Aversive Learning and Memory

Title: Chronic activation of NMDA-type glutamate receptors in the lateral amygdala after consolidation of associated fear-memory accelerates extinction but not forgetting

Authors: *J. TACHIKAWA[1], T. HATA[2];
[1]Doshisha Univ. - Kyotanabe Campus 1-3, Kyoto, Japan; [2]Psychology, Doshisha Univ., Kyotanabe, Japan

Abstract: It has already been shown that chronic inhibition of NMDA-type receptors improves retention of consolidated spatial-memory (Sachser et al., 2016; Shinohara et al., 2014, 2018; Villarreal et al., 2002). Conversely, can the activation of these receptors after consolidation decrease memory retention? If so, this could lead to the development of new treatments for those who have traumatic memories. In this study, rats were given paired or unpaired presentation of tone (CS) and electric foot-shock (US). From 24 hours after the presentation, very low concentration of NMDA solution or aCSF was chronically administered with osmotic pumps to the lateral amygdala. Twenty-four hours before the test, the administration was terminated by cutting the tubes connecting the pumps and canulae. Fourteen days after CS and US presentation, we executed the test session. In the test session, we presented CS alone and measured fear responses expressed as conditioned suppression of water drinking behavior. In the paired groups, there was no difference in fear response between the aCSF- and NMDA-treated groups in the early phase of the test. However, in the middle phase of the test, the NMDA group showed weaker fear responses than the aCSF group. In the late phase of the test, both groups showed extinction and the same level of fear response. In the unpaired groups, both the aCSF and NMDA groups consistently exhibited similar fear responses throughout the test. In addition, the weaker fear responses were shown in the unpaired group than in the paired group. From these results, we conclude that chronic activation of NMDA-type receptors in the lateral amygdala after the consolidation of associated fear-memories accelerates the progression of extinction, but not forgetting, of the memories. These results may lead to the development of pharmacotherapy as an adjunct to psychotherapy for those who have traumatic memories.

Disclosures: **J. Tachikawa:** None. **T. Hata:** None.

Poster

PSTR162. Mechanisms of Fear and Extinction Memory Modification

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR162.07/JJ20

Topic: G.04. Emotion

Support: NIH (R01 NS106915)
VA (IO1 BX003893)
The Brown Foundation

Title: Activation of cerebellar Purkinje cells disrupts the reconsolidation of associative fear memory

Authors: *G. KOGIAS[1,2], S. LIU[3,2];
[1]LSU Hlth. Sci. Ctr. Cell Biol. & Anat., NEW ORLEANS, LA; [2]Southeast Louisiana VA Healthcare Syst., New orleans, LA; [3]Cell biology and Anat., Louisiana State Univ. Hlth. Sci. Ctr., NEW ORLEANS, LA

Abstract: Cerebellar activity is critical for associative fear memory formation. Our recent study demonstrated that fear conditioning reduces the content of 2-Arachidonoylglycerol, an endocannabinoid in cerebellar lobules V/VI and this is required for memory consolidation. Once consolidated, memory can be modified. Following reactivation of the learned memory, the original memory becomes transiently labile and undergoes re-stabilization (a process called "reconsolidation"). This provides a window of opportunity that can strengthen or weaken the memory. Cerebellar activity is critical for memory reconsolidation as inhibition of protein synthesis in the cerebellar vermis during the reconsolidation period impairs memory recall. Here, we determined whether the activity of cerebellar Purkinje cells controls the reconsolidation of fear memory, using a Pavlovian fear conditioning paradigm (FC). We have previously shown that in vivo pharmacogenetic activation of the Gq pathway (with Gq coupled Designer Receptors Exclusively Activated by Designer Drugs) in Purkinje cells (PCs) after fear conditioning reduced the cued fear memory retention. We therefore subjected L7::Gq(+)-DREADD mice to FC (tone + shock) and then a re-activation stimulus (tone alone) 10 days later in a novel context, and they received J60, a DREADD receptor agonist immediately afterwards. 3 days after the reactivation of the fear memory, we measured the retention of the fear memory. L7::Gq(+)-DREADD mice exhibited a reduced freezing response to tones relative to two control groups (L7::Gq(-)-DREADD mice administered with J60 and saline-injected L7::Gq(+)-DREADD mice). Thus, activation of GqDREADD in PCs disrupted memory reconsolidation. To confirm that the memory loss following activation of Gq in PCs, was caused by a reduction in original fear memory, mice were subjected to a reinstatement test. One day before testing the reinstatement of the fear memory, a single foot-shock was delivered to the mice as a reminder. Mice that expressed Gq-DREADD in PCs exhibited reduced freezing responses during the memory

reinstatement test compared with control groups, suggesting a loss of original fear memory. The effects of PC activation on the reconsolidation and the reinstatement of the fear memory were only observed in male L7::Gq(+)-DREADD mice, but not in females. Therefore, our results suggest that pharmacological activation of the Gq pathway in PCs selectively disrupts reconsolidation of fear memory in male mice.

Disclosures: **G. Kogias:** None. **S. Liu:** None.

Poster

PSTR162. Mechanisms of Fear and Extinction Memory Modification

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR162.08/JJ21

Topic: G.01. Fear and Aversive Learning and Memory

Title: Targeted memory reactivation during post-learning sleep affects fear learning with the alterations of dendritic spine plasticity

Authors: *Q. ZHENG[1], X. HU[2,3], C. S. LAI[1,3];
[1]Sch. of Biomed. Sci., [2]Dept. of Psychology, [3]State Key Lab. of Brain and Cognitive Sci., Univ. of Hong Kong, Hong Kong, China

Abstract: Targeted memory reactivation (TMR) is a method to manipulate memory consolidation by presenting conditioned cues during post-learning sleep. Although TMR in non-rapid eye movement (NREM) sleep generally improved learning outcomes on verbal and procedural memory tasks, TMR exhibited contradictory effects on fear memory in humans and rodents, and the underlying mechanism is unclear. In this study, we evaluated fear-induced freezing behavior and dendric spine plasticity in the frontal association cortex (FrA) to examine the effects of TMR under different conditions. We found that TMR during slow-wave sleep (SWS) enhanced fear memory, but TMR during non-slow wave of NREM sleep (NS) impaired fear memory. The opposite effects of TMR correlated with fear learning-induced dendritic spine plasticity in the FrA and with the slow oscillation-spindle couplings. In sum, our data showed that TMR during different substages of NREM sleep exerted opposite effects on fear memory consolidation.

Disclosures: **Q. Zheng:** None. **X. Hu:** None. **C.S. Lai:** None.

Poster

PSTR162. Mechanisms of Fear and Extinction Memory Modification

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR162.09/JJ22

Topic: G.01. Fear and Aversive Learning and Memory

Support: Internal Project Award from the University of Massachusetts

Title: Estrogen Modulation of Recent vs. Remote Fear Extinction Memory Recall in Female Rats

Authors: *C. HARTSGROVE[1,2], G. WALKER[1], K. YUKNEWICZ-BOISVERT[3], M. ASKLER[1], S. LEE[1], Z. G. MICHAELS[1], L. Y. MAENG[1];
[1]Developmental and Brain Sci., Univ. of Massachusetts Boston, Boston, MA; [2]Psychology, Univ. of Massachusetts Boston, Dorchester, MA; [3]Psychology, Wheaton Col. (MA), Wheaton, MA

Abstract: Anxiety and stress-related disorders, such as post-traumatic stress disorder, are reported to be more prevalent in women. However, the neurobiological mechanisms underlying sex differences in these disorders remain unclear. Neural circuits involved in fear responses are altered in individuals affected by stress-related disorders and can be modulated by sex steroid hormones such as estrogen. Fear extinction is the ability to inhibit or control a fear response after a conditioned stimulus is no longer present and is dependent on distinct fear network activity. Prior research has demonstrated that higher estrogen levels during extinction training facilitates extinction recall 24 hours later. The current project examined how long the enhancing effects of estrogen on extinction recall last and how this effect might be regulated by the ERK pathway and gut microbiome. Naturally cycling adult female Sprague-Dawley rats underwent a 3-day fear conditioning/extinction paradigm. On Day 1, all animals in estrus (a period of low circulating estrogen levels) completed habituation and conditioning. Extinction training took place on Day 2 while subjects were in metestrus. 30 minutes prior to extinction training, 7 rats were administered estradiol (15ug/kg), and 7 rats were administered a sesame oil vehicle. Recent extinction recall test took place on day 3. Remote extinction recall test took place 7-15 days after extinction training. Freezing behavior across all behavioral phases was manually scored by 3 blinded, trained raters and averaged as two-trial blocks. An independent-samples t-test revealed that estrogen-treated rats showed significantly lower freezing behavior compared to vehicle-treated rats during the recent recall memory test ($t(12) = -2.568$, $p = .012$) but not the remote recall test ($t(12) = -1.054$, $p = .156$). Administration of estrogen resulted in short-term facilitation of fear extinction memory consolidation but not long-term facilitation, as seen by lower average freezing behavior during recent extinction recall but not remote extinction recall. Further analyses are being conducted on how the gut microbiome and activation of the ERK pathway within the fear circuitry contribute to the effect of estrogen levels on fear extinction recall.

Disclosures: C. Hartsgrove: None. **G. Walker:** None. **K. Yuknewicz-Boisvert:** None. **M. Askler:** None. **S. Lee:** None. **Z.G. Michaels:** None. **L.Y. Maeng:** None.

Poster

PSTR162. Mechanisms of Fear and Extinction Memory Modification

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR162.10/JJ23

Topic: G.01. Fear and Aversive Learning and Memory

Support: PAPIIT Grant IN205622
CONACYT Grant CVU:1083434

Title: Nt-3 and the strengthening of memory

Authors: *M. BRIONES VIDAL, S. E. REYES-GARCÍA, M. L. ESCOBAR;
Facultad de Psicología, Univ. Nacional Autonoma de Mexico, Ciudad de México, Mexico

Abstract: NT-3 and the strengthening of memory
M. G. Briones-Vidal, S. E. Reyes-García and M. L. EscobarDivisión de Investigación y Estudios de Posgrado, Facultad de Psicología, Universidad Nacional Autónoma de México, 04510, México
Research on the molecular mechanisms underlying learning and memory has demonstrated that neurotrophins play a crucial role on the establishment of long-lasting synaptic modifications required for memory formation. NT-3, a member of the neurotrophin family, is a key modulator of synaptic transmission and connectivity, but its role on memory processes has been scarcely explored. In this regard, resent evidence suggest that NT-3 diminishes memory alterations observed in pathological conditions, mainly in hippocampus-dependent tasks. However, NT-3 and its high affinity receptor, TrkC, are widely expressed in neocortical areas, such as insular cortex, a region in the temporal lobe critical for sensory integration, acquisition, and retention of aversive memories. Here we explored the effect of NT-3 on the consolidation of the conditioned taste aversion (CTA), a learning paradigm in which an animal associates a novel taste with gastric malaise and where the participation of the IC is fundamental. Our findings show that NT-3 strengthens the CTA memory trace. These data provide evidence that NT-3 is a potent regulator of the consolidation of aversive memories in the insular cortex, strengthening a memory trace in the adult brain.
Key words: Neurotrophin-3, CTA, Memory enhancement, Trk, insular cortex

Disclosures: M. Briones Vidal: None. **S.E. Reyes-García:** None. **M.L. Escobar:** None.

Poster

PSTR162. Mechanisms of Fear and Extinction Memory Modification

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR162.11/JJ24

Topic: G.01. Fear and Aversive Learning and Memory

Support: CDMRP TP210230

Title: Sex differences in delayed fear incubation in mice

Authors: *M. LITTLEPAGE-SAUNDERS, M. RUSNAK, M. TSUDA, T. WU;
Uniformed Services Univ. of the Hlth. Sci., Bethesda, MD

Abstract: Post-traumatic stress disorder (PTSD) has a lifetime prevalence of 1 in 13 people. It is not fully understood why some are more vulnerable to developing PTSD whereas others are more resilient. Among those who develop PTSD, about a quarter have delayed-onset PTSD where onset of symptoms occur at 6 months or later after experiencing a traumatic event. While there is recognition of this type of PTSD, the underlying mechanism is poorly understood. The development of PTSD is more prevalent in women than men. However, previous preclinical and clinical studies have focused mainly on males. Thus, there lacks an understanding of this sex difference. To enhance our understanding of the causal mechanisms, we designed a study with mice to compare the effect of sex on delayed fear incubation. We expected the female mice to show increased fear responses that would suggest more chronic delayed fear incubation than the male mice. To test this, we evaluated distance travelled, freezing, and latency until the first freezing episode in adult male and female C57Bl/6J mice (7-8 weeks of age; n=8/group) using a delayed fear incubation behavioral assay. Animals were placed in a fear conditioning chamber for a 198 s acclimation period and then administered a single 2 s scrambled footshock of 1.5 mA via the metal grid floor. After an additional 60 s, animals were returned to their home cage. At specific time after shock application, animals underwent a fear recall test where the animals were placed back in the shock chamber for a total of 180 s without any additional footshocks and then returned to their home cage. The study was based on a 2X2 factorial design comprising of sex (male vs female) and recall time (1 day and 14 days post shock). The data showed an increasing freezing trend (p=0.08) at d 14 compared to their recall d 1 counterparts. There was no effect of time or sex on distance travelled or latency until the first freezing episode. Interestingly, female mice showed less freezing when compared to the male mice. Ongoing experiments will look more into circuitry-level changes by comparing neuronal activation through cFos expression in different brain regions that are a part of the limbic system and/or have been associated with fear: the amygdala, hippocampus, prefrontal cortex, and paraventricular nucleus.

Disclosures: M. Littlepage-Saunders: None. **M. Rusnak:** None. **M. Tsuda:** None. **T. Wu:** None.

Poster

PSTR162. Mechanisms of Fear and Extinction Memory Modification

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR162.12/JJ25

Topic: G.01. Fear and Aversive Learning and Memory

Support: SFB1280

Title: Comparing extinction learning and memory suppression using a multi-paradigmatic approach

Authors: *G. JACOB[1,2], S. LISSEK[4], T. OTTO[3], C. J. MERZ[3], A.-C. SCHMIDT[2], L. N. WOLSINK[3], G. WALDHAUSER[2], H. ZHANG[2], N. AXMACHER[2];
[2]Inst. of Cognitive Neuroscience, Neuropsychology, [3]Dept. of Cognitive Psychology, [1]Ruhr Univ. Bochum, Bochum, Germany; [4]Dept. of Neurol., BG Univ. Hosp. Bergmannsheil, Bochum, Germany

Abstract: Extinction learning and memory suppression are two different forms of memory inhibition. Impaired extinction learning and memory suppression have been implicated in anxiety and post-traumatic stress disorder. However, the overlap in terms of underlying cognitive mechanisms, or lack thereof, between them remains unclear. Therefore, we aimed to investigate the relationship between extinction learning and memory suppression using a multi-paradigmatic approach. We recruited 54 participants, each of whom was administered classical fear extinction (FE), predictive learning (PL), and think/no-think (TNT) tasks. The FE and PL tasks measured extinction learning, with and without a fear component respectively, whereas the TNT task measured suppression-induced forgetting (SIF). We performed a preliminary analysis with 20 participants, and conducted a correlation analysis between the accuracy ratings of the PL task during extinction training and SIF as measured by the TNT task. We calculated the Spearman correlation coefficient ($r = 0.196$) between extinction learning and SIF, indicating a weak, non-significant correlation. A power analysis conducted based on this correlation coefficient (effect size = 0.397; alpha = 0.05; power = 0.8) revealed a required sample size of 67 participants. The preliminary results indicate that participants who showed better extinction learning performance may be better at memory suppression (and vice versa). This finding lends credence to the possibility that similar cognitive processes may be recruited during extinction learning and memory suppression, and this cognitive overlap may represent a target area during therapy for disorders with impaired extinction learning and memory suppression abilities. In the future, we aim to conduct a magnetic resonance imaging study using the same three tasks to investigate the potential overlap between the neural signatures of memory inhibition processes.

Disclosures: G. Jacob: None. **S. Lissek:** None. **T. Otto:** None. **C.J. Merz:** None. **A. Schmidt:** None. **L.N. Wolsink:** None. **G. Waldhauser:** None. **H. Zhang:** None. **N. Axmacher:** None.

Poster

PSTR162. Mechanisms of Fear and Extinction Memory Modification

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR162.13/KK1

Topic: G.01. Fear and Aversive Learning and Memory

Support: Supported by Harvard University

Title: Post-reactivation amygdala connectivity changes underlie reconsolidation-updating in humans

Authors: *E. S. LAURENT[1], M. C. KROES[2], J. E. DUNSMOOR[3], L. DAVACHI[4], E. A. PHELPS[1];
[1]Harvard Univ., Cambridge, MA; [2]Donders Inst. for Brain, Cognition, and Behaviour, Radboud Univ. Nijmegen Med. Ctr., Nijmegen, Netherlands; [3]Psychiatry, Dell Med. School, Univ. of Texas at Austin, Austin, TX; [4]Psychology, Columbia Univ., New York, NY

Abstract: Threat conditioning research in rodents demonstrates that a reminder of a previously acquired threat cue can result in memory destabilization creating an opportunity to update the threat memory with precisely timed extinction training, thus eliminating the return of threat responses (Monfils et al., 2009). Although similar behavioral findings have been demonstrated in humans (Schiller et al., 2010), neural evidence of memory reactivation that may induce destabilization is lacking. We tested participants on three consecutive days using a simple discrimination threat conditioning paradigm. On Day 1 participants underwent threat acquisition. The following day, half of the participants received a reminder to reactivate the threat memory and, after a 10-minute rest period, all participants received extinction training. On Day 3 we tested for the recovery of threat responses. Task blocks and the reminder cue were preceded and followed by 4.5-min rest periods. Blood oxygenation level dependent (BOLD) signal was acquired throughout the tasks and interspersing rest periods and skin conductance responses (SCRs) to the conditioned stimuli were assessed. SCR results showed evidence for threat memory acquisition and threat extinction in all participants.
Replicating previous results (Schiller et al., 2010), on Day 3 only participants who did not receive the reminder cue prior to extinction training showed evidence of threat recovery ($F_{1,32}$=4.466, p=.042). To determine BOLD patterns that characterized threat memory reactivation, and potential destabilization, on Day 2, we examined connectivity changes with the amygdala during resting scans prior to and following the cue reminder, and compared these connectivity changes with rest scans during the same time periods in the group that did not receive the threat cue reminder. Not surprisingly, there were no pre-post amygdala connectivity changes in the no-reminder group. However, there was a change in amygdala BOLD connectivity in the reminder group. These findings provide a potential neural signature that underlies memory reactivation during threat memory reconsolidation-updating in humans.

Disclosures: E.S. Laurent: None. **M.C. Kroes:** None. **J.E. Dunsmoor:** None. **L. Davachi:** None. **E.A. Phelps:** None.

Poster

PSTR162. Mechanisms of Fear and Extinction Memory Modification

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR162.14/KK2

Topic: G.01. Fear and Aversive Learning and Memory

Support: DFG; German Research Foundation
DFG; SFB1280 – project number 316803389
DFG; ZL 59 4–1 and ZL 59 5–1

Title: Effects of Verbal Instructions on Fear Extinction and Extinction Retrieval in Patients with Anxiety Disorders

Authors: A. LIPP[1], C. J. MERZ[2], O. T. WOLF[2], ***A. ZLOMUZICA**[1];
[1]Behavioral and Clin. Neurosci., [2]Cognitive Psychology, Ruhr-Universität Bochum, Bochum, Germany

Abstract: Patients with anxiety disorders (AD) show delayed extinction of conditioned fear responses. Explicit instructions given to participants prior to extinction (to increase conscious information about the CS-UCS occurrence) have been shown to affect fear extinction. In healthy subjects, instructions about the CS-UCS contingency given prior to extinction learning accelerate fear extinction.
Here, we investigated to which extent fear extinction and extinction retrieval can also be modified in patients with anxiety disorders by using explicit instructions about the CS-UCS contingency prior to and after fear extinction learning. We present preliminary findings which indicate that providing explicit instructions leads to changes in the rate and strength of fear extinction learning and retrieval in AD patients relative to healthy controls. Our findings could be used to develop more effective exposure therapy protocols in patients with AD.

Disclosures: **A. Lipp:** None. **C.J. Merz:** None. **O.T. Wolf:** None. **A. Zlomuzica:** None.

Poster

PSTR162. Mechanisms of Fear and Extinction Memory Modification

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR162.15/KK3

Topic: G.01. Fear and Aversive Learning and Memory

Support: CRPP Synapse & Trauma, University of Zürich

Title: Impact of MMP9 inhibitors doxycycline and minocycline on human aversive memory consolidation

Authors: J. WEHRLI[1], Y. XIA[2], B. KLEIM[3], ***D. R. BACH**[4];
[1]Univ. of Zurich, [2]Dept. of Psychiatry, Psychotherapy, and Psychosomatics, Univ. of Zurich, Zürich, Switzerland; [3]Dept. of Psychology, Univ. of Zürich, Zürich, Switzerland; [4]Hertz Chair for Artificial Intelligence and Neurosci., Univ. of Bonn, Bonn, Germany

Abstract: Matrix metalloproteinase 9 (MMP9) signalling is required for synaptic consolidation in LTP models, and in non-human models of hippocampus-dependent memory. This might open an avenue for clinical application, to prevent stress-related disorder after psychological trauma. However, it is unclear whether (a) MMP9 signalling is also required for human memory consolidation, and (b) it is required in aversive memory models, which have yielded mixed evidence in rodents. Here, we address this question in 4 pre-registered randomised placebo-controlled trials (RCTs) using a single dose (200 mg) of the MMP-inhibiting tetracycline

antibiotics doxycycline and minocycline. RCT1 (N = 76) tested the broadband MMP inhibitor doxycycline against placebo in a delay cued fear conditioning task. When learning took place under the influence of doxycycline, memory retention after 7 days was reduced. However, this was not replicated in two hippocampus-dependent memory models: RCT2 (N = 97) with trace (15 s interval) fear conditioning found mixed evidence, and RCT3 (N = 100) with contextual fear conditioning found no evidence for a consolidation impairment. In contrast, RCT4 (N = 105) showed that minocycline, which has higher MMP9 specificity, reduced contextual fear retention after seven days, compared to placebo and compared to the doxycycline group in RCT3. Overall, doxycycline appears to be less suited for clinical translation, but minocycline might offer a promising alternative with notably better pharmacokinetics.

Disclosures: J. Wehrli: None. **Y. Xia:** None. **B. Kleim:** None. **D.R. Bach:** None.

Poster

PSTR163. Neural Circuits of Fear and Aversive Processing

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR163.01/KK4

Topic: G.01. Fear and Aversive Learning and Memory

Support: Fondazione Istituto Italiano Di Tecnologia

Title: Synaptic endocannabinoid signaling modulates the reciprocal functional connectivity between the prefrontal cortex and the noradrenergic locus coeruleus

Authors: A. LOCARNO[1], L. NAVA[1], N. BARSOTTI[2], A. CALTABIANO[1], I. MISEVICIUTE[1], Y. PELLOUX[1], F. BOI[3], M. VINCENZI[3], D. P. COVEY[4], Y. LI[5], F. E. GEORGES[6], L. BERDONDINI[3], J. F. CHEER[7], M. PASQUALETTI[2], ***R. TONINI**[1]; [1]Neuromodulation of Cortical and Subcortical Circuits, Fondazione Inst. Italiano Di Tecnologia, Genova, Italy; [2]Dept. of Biol., Universitá di Pisa, Pisa, Italy; [3]Microtechnology for Neuroelectronics, Fondazione Inst. Italiano Di Tecnologia, Genova, Italy; [4]Univ. of Maryland, Baltimore, MD; [5]Peking Univ., Beijing, China; [6]IINS-CNRS 5297, Bordeaux, France; [7]Anat. and Neurobio., Univ. of Maryland Sch. of Med., Baltimore, MD

Abstract: The prefrontal cortex (PFC) is a key brain region involved in higher-order cognitive functions, including cognitive flexibility, attentional processes, and emotional regulation. This brain region is densely innervated by several long-range neuromodulatory afferents, including noradrenergic inputs originating from the Locus Coeruleus (LC). The PFC, in turn, sends glutamatergic projections to the LC, suggestive of a functionally interconnected loop between these two regions. Whether the strength of PFC to LC (PFC→LC) synapses is modulated by locally-produced neuromodulators to tune norepinephrine (NE) release in the PFC and change PFC neural dynamics through long-range connectivity remains unclear. By combining rabies tracing, optogenetics, and neurophysiological approaches, we provided evidence of a monosynaptic PFC→LC connection that is modulated by endocannabinoids (eCB) through the

activity of the cannabinoid receptor type 1 (CB1R) expressed at PFC terminals. In vivo fiber photometry of the genetically encoded NE sensor $GRAB_{NE}$, and High-Density single-unit recordings with SiNAPS probes showed that optogenetic activation of PFC→LC terminals induces norepinephrine (NE) release in the PFC and changes in the evoked PFC neural dynamics. eCBs generated locally in the LC promote NE-mediated activity remodeling in the PFC, as indicated by changes in evoked PFC activity when a CB1R antagonist was administered in the LC during PFC→LC stimulation. In summary, our data not only demonstrate a monosynaptic descending PFC to LC input and supports the existence of a reciprocally connected PFC-LC network but also show how eCBs locally released *on-demand* induce neuronal activity remodeling in distal brain regions.

Disclosures: A. Locarno: None. **L. Nava:** None. **N. Barsotti:** None. **A. Caltabiano:** None. **I. Miseviciute:** None. **Y. Pelloux:** None. **F. Boi:** None. **M. Vincenzi:** None. **D.P. Covey:** None. **Y. Li:** None. **F.E. Georges:** None. **L. Berdondini:** None. **J.F. Cheer:** None. **M. Pasqualetti:** None. **R. Tonini:** None.

Poster

PSTR163. Neural Circuits of Fear and Aversive Processing

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR163.02/KK5

Topic: G.01. Fear and Aversive Learning and Memory

Support: 5R00MH123495

Title: The Role of Lateral Habenula Circuits in Approach/Avoidance Conflict

Authors: *J. PEREZ[1], C. MARÍA-RÍOS[2], C. BRAVO-RIVERA[3];
[1]UPR-RCM, Guaynabo, Puerto Rico; [2]Univ. of Puerto Rico Rio Piedras Campus, San Juan, PR; [3]Psychiatry and Anat. & Neurobio., Univ. of Puerto Rico Departments of Psychiatry and Anat. & Neurobio., San Juan, PR

Abstract: Balancing motivational drives, such as reward approach and threat avoidance, is crucial for making appropriate choices. Dysregulated motivational drives can lead to excessive reward approach or persistent threat avoidance, which are hallmarks of addiction and anxiety disorders, respectively. Characterizing motivation circuits that govern reward/avoidance conflict behavior is necessary to understand the underpinnings of these disorders. The lateral habenula (LHb) is a key mediator of motivation, and more recently it has been implicated in fear expression exclusively in conflict contexts (Velazquez-Hernandez & Sotres-Bayon 2021). Although much is known of the role of LHb in negative affect and reward seeking, it remains unclear how the habenula influences behavior during motivational conflict. Here, we adapted the platform-mediated avoidance task, such that thirsty mice can nose-poke a reward port for water reward and avoid a tone-signaled foot-shock by stepping onto a safety platform away from the reward port. This approach sets a salient dichotomous motivational conflict between approach

and avoidance for probing motivational circuits. We found that optogenetic or chemogenetic activation or silencing of LHb without neuronal-type specificity had no effect on approach or avoidance during a conflict challenge. Interestingly, optogenetic activation of a subset of LHb neurons expressing GAD2 markedly increased avoidance in a conflict challenge, whereas silencing LHb GAD2 neurons did not affect approach or avoidance. Furthermore, optogenetic activation of LHb induced real-time place aversion whereas activation of LHb GAD2 neurons had no effect. Non-specific optogenetic activation of LHb neurons increased social interaction with a same-sex stranger conspecific, but not with a mouse of the opposite sex. Finally, optogenetic, or chemogenetic manipulations of non-specific LHb neurons did not affect time spent on open arms in an elevated plus maze, or time spent near the center of an open field. These data highlight the importance of neuronal subpopulations in characterizing the role of LHb in motivational conflict.

Disclosures: **J. Perez:** None. **C. María-Ríos:** None. **C. Bravo-Rivera:** None.

Poster

PSTR163. Neural Circuits of Fear and Aversive Processing

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR163.03/KK6

Topic: G.01. Fear and Aversive Learning and Memory

Support: NRF- 2021R1C1C1006607
NRF-2022M3E5E8018421
NRF-2022R1A2C2009265
NIH Grant MH099073
NRF-2021R1A6A3A01086526

Title: Distinct anterior insular circuits bi-directionally modulate conditioned fear in mice

Authors: *S. PARK[1], Y. HUH[2], E. KIM[3], J. J. KIM[3], J. CHO[4];
[1]Ewha Women's Univ., Seoul, Korea, Republic of; [2]Catholic Kwandong Univ., Catholic Kwandong Univ., Incheon, Korea, Republic of; [3]Univ. of Washington, Univ. of Washington, Seattle, WA; [4]Ewha Womans Univ., Ewha Womans Univ., Seoul, Korea, Republic of

Abstract: Abnormal activity in the anterior insular cortex (aIC) has been suggested to be linked to fear related disorders such as post-traumatic stress disorder (PTSD). However, detailed neuronal activity and the circuitry of the aIC involved in regulating fear remain undetermined. By utilizing behavioral single-unit recording in mice, we found that the activity of some pyramidal neurons in the aIC was either positively or negatively correlated with freezing (fear) behavior triggered by conditioned stimulus (CS: tone). No such correlation between tone and aIC neuronal activity was found in control mice that did not have fear conditioning experience. Optogenetically increasing or decreasing the activity of aIC pyramidal neurons during CS presentation respectively increased and decreased fear response, supporting that aIC neuronal

activity regulates conditioned fear response. In addition, aIC projections to different brain areas were confirmed to bidirectionally modulate fear; optogenetic activation of aIC projection to the amygdala increased fear behavior while activation of the aIC projection to the medial thalamus decreased fear behavior. Neural tracing revealed that the non-overlapping neuronal populations in the aIC projected to either the amygdala or the medial thalamus. Overall, our data support that the balance of positive and negative aIC neuronal activity is correlated with behavior and that aIC outputs collaborate to intricately modulate fear behavior.

Disclosures: **S. Park:** None. **Y. Huh:** None. **E. Kim:** None. **J.J. Kim:** None. **J. Cho:** None.

Poster

PSTR163. Neural Circuits of Fear and Aversive Processing

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR163.04/KK7

Topic: G.01. Fear and Aversive Learning and Memory

Support: 1F31MH125569-01A1
NIH R01 DA047443
Boettcher Foundation

Title: Vta to lateral habenula circuitry mediates the non-associative behavioral sequelae of inescapable stress experience

Authors: *D. MCGOVERN[1], H. MILLS[3], D. H. ROOT[2];
[1]Psychology & Neurosci., [2]Univ. of Colorado Boulder, Univ. of Colorado Boulder, Boulder, CO;
[3]3072 5th Street, NIDA, Baltimore, MD

Abstract: Ventral Tegmental Area glutamate neurons (VGluT2+) are significantly activated by inescapable stress and the activation of this population is necessary for the development of non-associative behavioral changes in male and female mice. The LHb forms bi-directional connections to the VTA and mediates aversive behaviors. We recently established that the VTA VGluT2+ → LHb pathway is significantly recruited by inescapable stress. In these experiments we evaluated the role of the LHb during escapable/inescapable stress and during post stress behavior, as well as the necessity of the VTA → LHb circuit for the development of behavioral changes induced by inescapable stress.Population level changes in intracellular calcium as well as glutamate and GABA input to LHb VGluT2+ neurons were measured using fiber photometry in combination with GCaMP6m, iGluSnFR, and iGABASnFR. GCaMP and iGluSnFR signaling was significantly increased by the presence of either escapable or inescapable stress, while GABASnFR signaling was significantly reduced in both conditions. During post stress foot-shock elicited fear, LHb GCaMP remained significantly elevated within the inescapable stress condition compared to the escapable stress condition. Glutamate input to LHb VGluT2+ neurons was significantly increased in both conditions while GABAergic input to LHb was reduced. GABASnFR signaling in the inescapable stress condition was significantly less than the

escapable stress condition. Next, we evaluated the role of 1) VTA VGluT2+ → LHb and 2) VTA VGluT2+/VGAT+ → LHb circuits in the development of non-associative behavioral changes following inescapable stress.These circuits were optogenetically inhibited during tail shock inescapable stress and male mice were evaluated for changes in social and fear behavior while female mice were evaluated for changes in exploratory behavior 24 hours later. Optogenetic inhibition of both the global VGluT2+ and the VGluT2+VGaT+ co-expressing sub-circuit prevented the development of social, fear, and exploration changes within male and female mice. This work establishes the role of LHb VGluT2+ neurons and their midbrain circuitry in the processing of inescapable stress stimuli and the development of non-associative behavioral changes following stress.

Disclosures: D. McGovern: None. **H. Mills:** None. **D.H. Root:** None.

Poster

PSTR163. Neural Circuits of Fear and Aversive Processing

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR163.05/KK8

Topic: G.01. Fear and Aversive Learning and Memory

Support: NSERC Discovery Grant 506730
CIHR 507489

Title: Characterization of the neural machinery for context-associated escape behaviours

Authors: *Y. HONG[1], J. BANG[1], A. K. BRINK[2], J. S. DIN[3], H. CHANG[3], J. KIM[2,1];
[1]Cell and Systems Biol., [2]Psychology, [3]Univ. of Toronto, Toronto, ON, Canada

Abstract: In nature, prey animals must constantly assess their environment to survive from nearby predators. In response to threatening stimuli, animals must display innate defensive responses including freezing, fleeing, and fighting. While recent studies highlight the neural mechanisms underlying instinctive escape behaviours, surprisingly little attention has been given to how environmental cues drive the control of these innate behaviours. Furthermore, it remains unclear how cell-type-specific projections differentially mediate innate and context-dependent innate defensive responses. Here, we demonstrate that the hippocampal projections to the anterior hypothalamic nucleus (AHN) mediate context-associated innate defensive behaviours. Furthermore, this work provides preliminary evidence which suggests unique roles for GABAergic and CaMKIIα^+ AHN neurons in mediating innate escape behaviours. Through anatomical tracing experiments, we demonstrate that the AHN is the only brain region within the medial hypothalamic defense system which receives input from the hippocampus (HPC). Subsequently, using fiber photometry, we found that GABAergic and CaMKIIα^+ AHN neurons show distinct dynamic responses to predators and contexts associated with predators. Moreover, optogenetic studies suggest unique roles for GABAergic and CaMKIIα^+ AHN neurons in mediating defensive behaviours. Together, these results provide strong preliminary evidence that

GABAergic and CaMKIIα^+ AHN neurons utilize projections from the HPC to mediate both innate escape behaviours and context-associated innate defensive behaviours.

Disclosures: **Y. Hong:** None. **J. Bang:** None. **A.K. Brink:** None. **J.S. Din:** None. **H. Chang:** None. **J. Kim:** None.

Poster

PSTR163. Neural Circuits of Fear and Aversive Processing

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR163.06/KK9

Topic: G.01. Fear and Aversive Learning and Memory

Support: NIH 5P01AI073693-13

Title: Long sepsis leads to impaired network encoding of fear

Authors: *J. J. STROHL, J. CARRIÓN, P. T. HUERTA;
Feinstein Inst. for Med. Research, Northwell Hlth., Manhasset, NY

Abstract: Understanding the long-term effects of sepsis has become critically important. As survival has increased, there is an ever-growing population of patients who live with long-term sequalae. Survivors frequently suffer long-lasting cognitive impairment, brain fog, and emotional imbalance, we call this condition long sepsis (LS). Mouse models of LS have shown that the brain, and in particular, the hippocampus is affected, and mice display impaired contextual fear conditioning. Our goal is to elucidate whether LS mice have functional abnormalities across the brain hubs that encode fear memory, comprising the hippocampus (HPC), basolateral amygdala (BLA) and prefrontal cortex (PFC). Septic shock was modeled by cecal-ligation-and-puncture in male C57BL/6 mice (50% lethality); controls (CON) underwent sham surgery. To specifically study long sepsis, we allowed a period of 6 weeks for recovery from initial shock. Following this extended period, mice were subjected to trace-fear-conditioning (TFC) paired with either positron emission tomography (PET) or electrophysiological recording. TFC consists of a 3-day paradigm where mice are familiarized to the chamber on day 1, presented with a series of paired tones and foot-shocks on day 2, and tested for context or tone memory on day 3. Mice undergoing TFC with PET were injected with (18)F-fluorodeoxyglucose (FDG) 25 min prior to behavioral testing, and subsequently placed onto the PET scanner. Mice undergoing recordings were implanted with multi-electrode arrays directed to the HPC, BLA and PFC (1 week prior to TFC). We took the difference in FDG standard uptake values (SUV) from the fear sessions (context memory - familiarization) and found that CON mice have a dramatic increase in BLA and PFC coupled with a decrease in the dorsal HPC. Strikingly, LS mice have significantly smaller changes, as measured by SUV difference, in BLA, PFC, and dorsal HPC. Neural recordings revealed altered oscillations in LS mice, such as persistently elevated powers in the theta and gamma bands, and altered theta-gamma coupling during fear memory sessions. We conclude that neural networks which encode fear are severely altered in LS mice. Our PET

studies have shown a diminished response of the brain substrate for TFC. Neural recordings revealed that oscillatory patterns, critical for coordination across brain regions, are disrupted in long sepsis, reducing the ability of the brain to recall and maintain an appropriate fear state.

Disclosures: J.J. Strohl: None. **J. Carrión:** None. **P.T. Huerta:** None.

Poster

PSTR163. Neural Circuits of Fear and Aversive Processing

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR163.07/KK10

Topic: G.01. Fear and Aversive Learning and Memory

Support: KBRI funded by Ministry of Science and ICT, 23-BR-03-03
NRF of Korea funded by the Ministry of Education, NRF-2020R1A6A3A1307717711
NRF of Korea funded by the Ministry of Education, NRF-2022M3E5E8081182

Title: Parietal-frontal pathway controls relapse of fear memory in a novel context

Authors: *B. JOO[1,2], J. KOO[1,2];
[1]Korea Brain Res. Inst., Daegu, Korea, Republic of; [2]Dept. of Brain Sci., Daegu Gyeongbuk Inst. of Sci. and Technol. (DGIST), Daegu, Korea, Republic of

Abstract: The ability to process contextual stimuli to adapt rapidly to novel circumstances is fundamental for survival, but maladaptive information processing can cause fear-related disorders. However, it remains unknown how novel contextual stimuli are associated with fear. We demonstrate that the posterior parietal cortex (PPC) to the anterior cingulate cortex (ACC) governs the relapse of extinguished fear memories in a novel context. We observed enhanced populational calcium activity in the ACC neurons that received projections from the PPC (PPC-ACC) and increased synaptic activity in the BLA-projecting PPC-ACC neurons upon renewal in a novel context, where excitatory postsynaptic currents amplitudes increased but inhibitory postsynaptic current amplitudes decreased. In addition, we found that parvalbumin (PV)-expressing interneurons (PPC-ACC^{PV}) control novel context-dependent fear renewal, which was blocked by the chronic administration of fluoxetine, a first-line pharmacotherapy for post-traumatic stress disorder. Our findings highlight the PPC-ACC pathway as a potential therapeutic target for fear-related disorders.

Disclosures: B. Joo: None. **J. Koo:** None.

Poster

PSTR163. Neural Circuits of Fear and Aversive Processing

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR163.08/KK12

Topic: G.01. Fear and Aversive Learning and Memory

Support: ERC Consolidator Grant
FRM (Fondation Recherche Médicale) : FDT202304016643
FRM (Fondation Recherche Médicale) : FDT201805005887

Title: Proximal and distal threat elicit opposing brain-body states during freezing

Authors: *B. MAHÉO[1], S. BAGUR[2], K. BENCHENANE[1];
[1]ESCPI Paris, Paris, France; [2]Inst. de l'Audition, Paris, France

Abstract: Fear encompasses multiple adaptive responses to different threat levels. In rodents, freezing is the dominant behavior. Since it is primarily characterized by immobility, freezing is considered a homogeneous state despite the fact that it appears in fear protocols eliciting very different threat levels (ex: tone/context conditioning, visual looming, predatory odor). This led us to ask if freezing is a more diverse set of states, that can be revealed by monitoring not just movement but also somatic and neurological state by which animals could covertly adapt to different threats.To address this issue, we recorded multiple cardio-respiratory variables, single units in the prefrontal cortex (PFC) and LFP in the hippocampus (HPC) while mice freely explored a U-shaped environment in which they received aversive shocks in one arm but were safe from shocks in the other. Mice froze in both arms, allowing us to compare freezing episodes proximal and distal to threat. Proximal to threat, cardio-respiratory state is activated (fast respiratory and heart rate, low heart-rate variability) with slow HPC theta and PFC single units entrained to breathing. Distal to threat, cardio-respiratory activity is reduced with numerous HPCal sharp-wave ripples (0.6/s) during which PFC replays post-shock activity. All differences, including PFC activity, were sufficiently robust to decode the animal's distance to the shock zone in real time.
We tested whether agents known to impact panic and anxiety, classically associated with high and low threat levels, specifically modify the two freezing types. Panicogenic dPAG stimulation induced and panicolytic (chronic fluoxetine) reduced proximal freezing, whereas an anxiogenic (nicotine) induced distal freezing, with more mixed results for anxiolytic (diazepam) administration.
Therefore, despite identical overt behavior, mice freeze close to threat in an aroused, panic-like state, compatible with flight preparation, but shift to a tranquilized, anxiety-like state dominated by internally-driven activity when threat is distal. The identification of these novel freezing types differing by somatic, neurophysiological and pharmacological profiles should allow this widely-studied behavior to be re-evaluated according to level of threat.

Disclosures: B. Mahéo: None. **S. Bagur:** None. **K. Benchenane:** None.

Poster

PSTR163. Neural Circuits of Fear and Aversive Processing

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR163.09/KK13

Topic: G.01. Fear and Aversive Learning and Memory

Title: Non-canonical cortico-thalamic dynamics gate avoidance decisions

Authors: *J. O'MALLEY[1,2], J. MA[2], M. KREIKER[2], Y. LENG[2], M. A. PENZO[2];
[1]NIMH, NIH, Bethesda, MD; [2]Natl. Inst. of Mental Hlth., Bethesda, MD

Abstract: The cortex exerts top-down control of the thalamus by either enhancing or suppressing incoming sensory signals to coordinate a behavioral response. The prelimbic area (PL) of the prefrontal cortex is critical for selection of avoidance behaviors. Yet it is unclear how the PL exerts top-down control over the thalamus to shape the selection of avoidance behaviors. Recent work has shown that the paraventricular nucleus of the thalamus (PVT), a part of the limbic system and has been shown to receive PL input, is integral for driving avoidance behavior. To assess if the PVT projecting PL neurons (PL-PVT) shapes the selection of avoidance behaviors, we used a 2-way active avoidance (2AA) paradigm. Briefly, mice were placed in a 2 chambered box and learned to avoid a footshock by shuttling to the opposite chamber in response to a warning signal (WS). Using fiber photometry, we recorded the activity of PL neurons during 2AA and found that PL-PVT showed bidirectional responses with increased activity to the WS and reduced activity when initiating shuttling to avoid or escape the footshock. The reduction in activity in PL-PVT is at odds with findings that PVT activity increases during avoidance, suggesting that PL might couple to PVT via disinhibitory mechanisms. Given that the PVT lacks interneurons, inhibition must come from elsewhere. One candidate for mediating this effect is the thalamic reticular nucleus (TRN), a GABAergic nucleus that solely targets the thalamus and receives cortical input that results in strong thalamic inhibition. To determine if the PL exerts a largely inhibitory effect over the PVT via the TRN, we used *ex vivo* slice electrophysiology with optogenetics and found that the PL sends weak excitatory inputs to the PVT whereas the TRN receives strong excitation from the PL and robustly inhibits the PVT. Next, using fiber photometry, we found that PL-TRN and TRN-PVT had bidirectional responses mirroring PL-PVT. Importantly, responses to the WS only occurred during avoidance trials, suggesting that the brief activity during the WS and subsequent reduction shapes the expression of avoidance. Indeed, optogenetic excitation mimicking the brief increase in activity increased avoidance. A key feature of thalamic cells is rebound activity following inhibition. The brief activation and reduction of TRN may lead to increased activity in PVT as observed in a fiber photometry experiment, suggesting a timing mechanism through coordinated disinhibition and subsequent excitation via the PL. Taken together, our findings identify a novel circuit (PL-TRN-PVT) that is critical for the expression of avoidance.

Disclosures: J. O'Malley: None. **J. Ma:** None. **M. Kreiker:** None. **Y. Leng:** None. **M.A. Penzo:** None.

Poster

PSTR163. Neural Circuits of Fear and Aversive Processing

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR163.10/KK14

Topic: G.01. Fear and Aversive Learning and Memory

Support: Z01ES090089

Title: Cholinergic Signaling in the ventral subiculum mediates avoidance response to threats

Authors: *S. WANG[1], J. L. YAKEL[2];
[2]Neurobio. Lab., [1]NIEHS, RTP, NC

Abstract: The ventral subiculum (vSub) serves as a crucial hub for information exchange between the ventral hippocampus (vHipp) and brain regions implicated in threat learning, such as the basal lateral amygdala (BLA). The vSub circuitry is regulated by cholinergic inputs from the basal forebrain. Our previous studies have revealed that vSub acetylcholine (ACh) levels respond to threatening stimulus like foot-shock and conditioned sound cue. However, the precise contribution of ACh in processing the threat info remains elusive. This prompts us to address questions regarding the role of cholinergic signaling: Does it solely convey sensory information? Does ACh respond to specific types of threats? Does ACh also participate in determining animal reactions to the threat? And what are the potential mechanisms underlying this process? To better understand the function of ACh signaling in threat response, we designed a behavioral task battery and systematically compared the dynamic change of the vSub ACh signaling at various stages of threat processing. In this study, we expressed a genetically encoded ACh sensor ($GRAB_{ACh3.0}$) in the vSub and used fiber photometry system to monitor the dynamic change of ACh levels in real time. Firstly, we found that vSub ACh levels increase in response to different types of threatening stimuli, including foot-shock, hand looming and tail-picking, while the mice also showed escaping behavior to these stimuli. Secondly, in a light-dark shuttle box test, we observed significant rise of ACh levels when the mice transitioned from the light box to the dark whereas ACh levels decreased when animals moved the other away. Similarly, a slight increase of ACh level is also observed in the elevated plus maze task when the mice moved to the closed arms. Notably, all ACh level changes occurred prior to the initiation of movements. Moreover, we identified an oscillatory pattern in vSub ACh dynamic when the mice were in the fear conditioning box following foot-shock, and the rising phase of ACh dynamics was paired with the transition from the freezing state to the moving state. Similar phenomena were also observed on mice injected with nicotine i.p. Taken together, these results support our hypothesis that ACh signaling in the vSub underlies the induction of avoidance response to threat, potentially involving a nicotinic ACh receptor-mediated pathway. Further experiments employing nicotinic AChR antagonists will be performed to confirm this hypothesis. By elucidating the intricate relationship between vSub ACh signaling and threat processing, our study provides insights into the neural mechanisms that govern behavioral responses to threatening stimuli.

Disclosures: S. Wang: None. **J.L. Yakel:** None.

Poster

PSTR163. Neural Circuits of Fear and Aversive Processing

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR163.11/KK15

Topic: G.01. Fear and Aversive Learning and Memory

Support: NIMH DP2 MH122399
NIHM R01 MH120162
Brain Research Foundation Award
Klingenstein-Simons Fellowship
NARSAD Young Investigator Award
McKnight Memory and Cognitive Disorder Award
One Mind-Otsuka Rising Star Research Award
Hirschl/Weill-Caulier Award
Mount Sinai Distinguished Scholar Award
Friedman Brain Institute Award
NIHM K99 MH131792
BBRF Young Investigator Award
NINDS F32 NS116416
AES Predoctoral Research Fellowship
NINDS R01 NS116357

Title: Dissociable contributions of the amygdala and ventral hippocampus to stress-induced changes in defensive behavior

Authors: *P. SOMPOLPONG[1], Z. T. PENNINGTON[4], A. LABANCA[5], S. D. ABDEL-RAHEIM[1], Z. CHRISTENSON WICK[1], Y. FENG[6], Z. DONG[2], T. FRANCISCO[7], L. CHEN[3], S. L. FULTON[8], I. S. MAZE[9], R. L. CLEM[10], T. SHUMAN[9], D. J. CAI[11];
[1]Neurosci., [3]Icahn Sch. of Med. at Mount Sinai, [2]Icahn Sch. of Med. at Mount Sinai, New York, NY; [4]Neurosci., [5]Icahn Sch. of Med. At Mount Sinai Grad. Training Program In Neurosci., [6]Icahn Sch. of Med. at Mount Sinai, [7]Icahn Sch. of Med. At Mount Sinai Grad. Training Program In Neurosci., New York, NY; [8]Columbia Univ., New York City, NY; [9]Neurosci., Icahn Sch. of Med. At Mount Sinai, New York, NY; [10]Neurosci., Mount Sinai Sch. of Med., New York, NY; [11]Mount Sinai, New York, NY

Abstract: Associative memories resulting from traumatic stress are the dominant target of trauma-based therapies. However, traumatic stress can produce an array of long-lasting changes in fear and defensive behaviors, including increased anxiety-like behavior and heightened reactions to novel aversive events, in addition associative fear memories. Notably, these behaviors are poorly correlated, suggesting they may reflect distinct defensive processes. From a clinical perspective, if these phenotypes are biologically dissociable, they may need to be independently targeted. To address this possibility, we combined pharmacological and chemogenetic methods with a behavioral protocol in which mice experience a severe stressor and a week later are exposed to a battery of fear and anxiety-like behavior tests. As expected, we found that blocking trauma-induced protein synthesis in the basolateral amygdala (BLA) and ventral hippocampus (vHC) reduced subsequent associative fear of the trauma environment.

However, blocking protein synthesis in the BLA profoundly mitigated heightened responses to novel aversive events but was without effect on anxiety-like behavior. Conversely, blocking protein synthesis in the vHC attenuated changes in anxiety-like behavior but was without effect on subsequent enhanced responding to novel aversive events. In line with our protein synthesis results, we found neuronal activity of the BLA and vHC differentially support the expression of these same defensive behaviors. Collectively, these results indicate that trauma-induced plasticity in BLA is critical for heightened reactions to novel aversive events while trauma-induced plasticity in vHC is critical for increased anxiety-like behavior. Moreover, they indicate that associative fear memories are dissociable from changes in anxiety-like behavior and stress sensitivity following trauma. We are now pursuing pathway-specific chemogenetic strategies to define the downstream effectors through which the BLA and vHC support these trauma-induced defensive behavior changes.

Disclosures: P. Sompolpong: None. **Z.T. Pennington:** None. **A. LaBanca:** None. **S.D. Abdel-Raheim:** None. **Z. Christenson Wick:** None. **Y. Feng:** None. **Z. Dong:** None. **T. Francisco:** None. **L. Chen:** None. **S.L. Fulton:** None. **I.S. Maze:** None. **R.L. Clem:** None. **T. Shuman:** None. **D.J. Cai:** None.

Poster

PSTR163. Neural Circuits of Fear and Aversive Processing

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR163.12/KK16

Topic: G.01. Fear and Aversive Learning and Memory

Support: NIMH DP2 MH122399
NIMH R01 MH120162
Brain Research Foundation Award
Klingenstein-Simons Fellowship
NARSAD Young Investigator Award
McKnight Memory and Cognitive Disorder Award
One Mind-Otsuka Rising Star Research Award
Hirschl/Weill-Caulier Award
Mount Sinai Distinguished Scholar Award
Friedman Brain Institute Award
NIMH K99 MH131792
BBRF Young Investigator Award

Title: Unbiased identification of a novel hypothalamic nucleus that regulates persistent fear and anxiety states after severe stress

Authors: *Z. PENNINGTON[1], P. SOMPOLPONG[3], A. LABANCA[2], S. D. ABDEL-RAHEIM[4], A. SMITH[5], Z. DONG[6], R. L. CLEM[7], P. J. KENNY[8], D. J. CAI[9];
[2]Icahn Sch. of Med. At Mount Sinai Grad. Training Program In Neurosci., [1]Icahn Sch. of Med.

At Mount Sinai Grad. Training Program In Neurosci., New York, NY; [3]2025 1st Ave, Icahn Sch. of Med., New York, NY; [4]Icahn Sch. of Med. At Mount Sinai Grad. Training Program In Neuroscience, New York, NY, United States, New York, NY; [5]Med. Univ. of South Carolina (MU Neurosci. Inst. - Grad., Charleston, SC; [6]Icahn Sch. of Med. at Mount Sinai, New York, NY; [7]Mount Sinai Sch. of Med., Mount Sinai Sch. of Med., Brooklyn, NY; [8]Icahn Sch. of Med. At Mount Sinai, Icahn Sch. of Med. At Mount Sinai, New York, NY; [9]Mount Sinai, New York, NY

Abstract: Acute severe stress is able to produce lasting enhancements of fear and anxiety, promoting a range of psychiatric illnesses. How this sensitization is instantiated in the nervous system, however, remains unclear. To address this issue, we first utilized unbiased whole-brain imaging of immediate-early genes in mouse brains to examine how a severe stressor is able to enhance subsequent responses to a novel stressor a week later. Among the observed changes, we found that the novel stressor led to robust hyperactivation of the anterior hypothalamic nucleus (AHN) in previously stressed animals. Notably, despite the AHN's rich interconnections with stress and defensive circuitry, its functional role has been explored little. To assess the causal contribution of the AHN to persistent fear and anxiety-like states after severe stress, we characterized the role of GABAergic and glutamatergic AHN neurons. We found that chemogenetic inhibition of AHN GABAergic neurons reduced associative fear of contextual cues previously associated with a severe stressor, and also reduced associative fear responses to subsequent aversive events. Conversely, stimulation of these neurons enhanced associative responding to subsequent aversive events, suggesting these neurons bidirectionally control associative fear. Corroborating this finding, using miniscope imaging of calcium activity, we found GABAergic AHN neurons respond acutely to multiple aversive events during associative learning. However, neither their inhibition or excitation influenced anxiety-like behavior in the light-dark test, highlighting a specific role of GABAergic neurons in defensive behavior, likely associative stress responses. To the contrary, manipulation of glutamatergic neurons within the AHN produced a very different pattern of results, with inhibition producing a profound reduction in anxiety-like behaviors. Collectively, these results highlight the AHN as a critical node regulating stress-induced changes in defensive behavior, and suggest GABAergic and glutamatergic neurons may contribute to distinct microcircuits within the AHN. We are now pursuing pathway-specific chemogenetic strategies to define the upstream regions that the AHN interacts with to support these trauma-induced defensive behavior changes.

Disclosures: Z. Pennington: None. **P. Sompolpong:** None. **A. LaBanca:** None. **S.D. Abdel-Raheim:** None. **A. Smith:** None. **Z. Dong:** None. **R.L. Clem:** None. **P.J. Kenny:** None. **D.J. Cai:** None.

Poster

PSTR163. Neural Circuits of Fear and Aversive Processing

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR163.13/KK17

Topic: G.01. Fear and Aversive Learning and Memory

Title: Neural mechanisms of Pathogen-Threat induced Social Avoidance

Authors: *F. MALTESE, C. BELLONE;
Basic Neurosciences, Univ. of Geneva, Geneva, Switzerland

Abstract: COVID-19 pandemic impacted social life by forcing “social distancing”. This measure has not only severely disrupted daily social interactions, but also led to an increase in mental health problems. As a result, understanding the neurobiological mechanisms underlying pathogen-threat induced social avoidance (PTSA) has become increasingly important. Studies on rodents have been proved valuable in exploring PTSA. Mice, for instance, innately avoid sick conspecifics, although little is known about the specific behavioral and circuit mechanisms underlying this response. We hypothesize that mice assign a lower reward value to sick compared to healthy conspecifics. To test this hypothesis, we developed a two-choice task allowing mice to decide between interacting with healthy or sick conspecifics. Our results revealed that mice spend more time interacting with healthy stimuli. We are currently assessing the neural mechanisms underlying this social decision-making process. The Nucleus Accumbens (NAc) is a key hub in the brain for translating motivation into action. We are testing the hypothesis that the activity of the NAc controls the decision to interact with healthy over the sick conspecifics. A thorough understanding of the behavioral and neural mechanisms underlying social-reward value assignment to pathogen threat could help us prevent the risks associated with social isolation.

Disclosures: **F. Maltese:** None. **C. Bellone:** None.

Poster

PSTR163. Neural Circuits of Fear and Aversive Processing

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR163.14/KK18

Topic: G.01. Fear and Aversive Learning and Memory

Title: Neuronal circuits underlying social threat recognition

Authors: *G. CASAROTTO, A. GEMELLI, C. BELLONE;
Basic Neurosci., Univ. de Geneve, Geneve, Switzerland

Abstract: Animals are constantly exposed to information from different environmental sources and must process them to make informed decisions and protect themselves from potential dangers. In this study, we used mice to investigate how the brain recognizes and responds to social threats, which ultimately enables adaptive behaviors. We first exposed mice to aggressive conspecifics and tested their ability to detect the threat and to learn to avoid it. We then studied the role of the basolateral amygdala to ventral striatum pathway in threat detection and subsequent behavioral adaptations. We were able to causally link the activity of this pathway to the mouse's ability to learn to recognize potential threats. Additionally, we identified input-specific forms of synaptic plasticity that instruct threat recognition and are induced by serotonin.

Our findings provide insight into the neural network associated with threat detection and the factors that contribute to adaptive behavior in response to social threats.

Disclosures: **G. Casarotto:** None. **A. Gemelli:** None. **C. Bellone:** None.

Poster

PSTR163. Neural Circuits of Fear and Aversive Processing

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR163.15/KK19

Topic: G.01. Fear and Aversive Learning and Memory

Support: R01MH117852

Title: Chemogenetic activation of infralimbic parvalbumin interneurons impairs extinction in male and female rats

Authors: *K. L. CRAYTON, A. N. BINETTE, H. BAYER, L. MELISSARI, S. O. SWECK, S. MAREN;
Texas A&M Univ. Neurosci. Inst. For Neurosci., College Station, TX

Abstract: Stress is a major contributor to many psychiatric disorders, particularly trauma- and anxiety-related disorders such as post-traumatic stress disorder (PTSD). Studies from our lab and others have shown that stress activates the basolateral amygdala (BLA), which in turn dampens activity in the infralimbic cortex (IL), a region of the medial prefrontal cortex (mPFC) that is critical for the reduction of learned fear (i.e., fear extinction). Although fear memory is durable and adaptive, fear extinction is easily disturbed. We propose that under high-stress conditions, the BLA has an inhibitory effect on the activity of IL principal neurons via inhibitory parvalbumin (PV) interneurons. Prior work in our lab has shown that footshock stress induces Fos expression in mPFC PV interneurons in IL. Here, we test the hypothesis that chemogenetic excitation of IL PV neurons will impair extinction. Male and female Long-Evans rats were injected with a viral vector in the IL to selectively drive the expression of an excitatory designer receptor exclusively activated by a designer drug (DREADD; AAV-S5E2-Gq-dTomato) in PV interneurons. After recovery, animals underwent a standard auditory fear conditioning procedure (5 tone-footshock trials). Twenty-four hours later, animals received systemic injections of either vehicle (VEH) or clozapine-N-oxide (CNO, 5 mg/kg, i.p.) followed by extinction training (45 tone-alone trials) in a novel context. All animals acquired extinction similarly, with an initial increase in CS-evoked freezing followed by a reduction in freezing across the session. However, CNO-treated male and female rats exhibited higher levels of freezing (normalized to baseline) compared to VEH- treated controls in an extinction retrieval test conducted 24 hours after extinction. These results demonstrate that chemogenetic excitation of IL PV interneurons impairs the encoding of long-term extinction memories in male and female rats. These data suggest that IL PV interneurons may be particularly sensitive to stress and contribute to stress-induced psychopathology and dysregulation of the mPFC.

Disclosures: **K.L. Crayton:** None. **A.N. Binette:** None. **H. Bayer:** None. **L. Melissari:** None. **S.O. Sweck:** None. **S. Maren:** None.

Poster

PSTR163. Neural Circuits of Fear and Aversive Processing

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR163.16/KK20

Topic: G.01. Fear and Aversive Learning and Memory

Support: R01MH065961

Title: Context fear memories are not necessary for renewal of extinguished fear in rats

Authors: *G. GARCIA, J. E. HASSELL, Jr, K. VASUDEVAN, V. VIERKANT, N. PHAM, C. V. TERCILLA, M. A. PARR, S. MAREN;
Texas A&M Univ. Neurosci. Inst. For Neurosci., College Station, TX

Abstract: Extinction learning is central to behavioral therapies for treatment of stressor- and trauma-related disorders including PTSD. We recently discovered that mPFC projections to the thalamic nucleus reuniens (RE), which projects strongly to the hippocampus (HPC), play a critical role in extinction learning and retrieval. After extinction, fear to the extinguished conditioned stimulus (CS) can relapse under a variety of conditions, including after an animal experiences the CS outside the extinction context—a phenomenon termed renewal. Additionally, pharmacological inactivation of the RE results in a relapse of extinguished fear after extinction. Behavioral studies suggest that renewal is not due to aversive properties of the test context, because it occurs in contexts that have never been paired with shock (e.g., ABC or ABB renewal). In Experiment (Exp 1), we examined whether RE promotes extinction retrieval by suppressing the retrieval of HPC-dependent fear memories. We predicted that preventing the formation of hippocampal context fear memories would reduce the relapse of extinguished fear associated with RE inactivation. To further explore this, in Exp 2 we examined whether antagonism of hippocampal NMDA receptors, which prevents the formation of context fear memories, would attenuate renewal of fear in the conditioning context. Animals received an intra-HPC infusion of saline or the NMDA receptor antagonist, AP5 (10 µg/µl, 0.3 µl per side) (n = 8 per group) and immediately underwent auditory fear conditioning (Exp 1 & Exp 2). The next day rats underwent fear extinction in a novel context, this was followed by an extinction retrieval test 24 hours later. In Exp 1, prior to the retrieval test, rats received intra-RE infusions of saline or muscimol. Consistent with prior findings, inactivation of the RE increased freezing to the CS in the extinction context. Additionally, prior hippocampal NMDA receptor antagonism attenuated the RE-inactivated induced increase in freezing. In Exp 2, animals treated with AP5 exhibited a robust deficit in freezing in the conditioning context, revealing that they failed to acquire a context-shock memory. Nevertheless, these rats showed robust renewal of extinguished responding to the CS in the conditioning context. These data are consistent with the hypothesis that RE is involved in suppressing inappropriate contextual fear memories (Exp 1) and that

renewal of fear does not require contextual fear memories (Exp 2). These results, consistent with other literature, suggest that only forms of relapse that involve direct context-shock associations (e.g., reinstatement) are attenuated by hippocampal NMDA receptor antagonism.

Disclosures: G. Garcia: None. **J.E. Hassell:** None. **K. Vasudevan:** None. **V. Vierkant:** None. **N. Pham:** None. **C.V. Tercilla:** None. **M.A. Parr:** None. **S. Maren:** None.

Poster

PSTR163. Neural Circuits of Fear and Aversive Processing

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR163.17/LL1

Topic: G.01. Fear and Aversive Learning and Memory

Title: Fear memory updating by exposure to an opposite-sex conspecific during retrieval: role of dopamine and oxytocin receptors

Authors: *A. ARELLANO PEREZ[1], A. KAUTZMANN SARTORI[2], S. MAREN[1], L. DE OLIVEIRA ALVARES[2];
[1]Psychological and Brain Sciences, Inst. for Neuroscience., Texas A&M University., College Station, TX; [2]Biophysics Dept., Univ. Federal do Rio Grande do Sul (UFRGS), Porto Alegre, Brazil

Abstract: It has been well established that a consolidated memory can be updated during the plastic state induced by reactivation. This updating process opens the possibility to modify maladaptive memory. In the present study, we evaluated whether fear memory could be updated to a less aversive level by incorporating hedonic information through its reactivation with the presentation of a conspecific. Here, male and female wistar rats were contextual fear conditioned and, during retrieval, a conspecific either female or male was presented as a social rewarding stimulus. We found that memory reactivation with conspecific reduced fear memory expression within-session, and in a session test, just males exposed to a female during reactivation reduced significantly levels of freezing, without presenting reinstatement or spontaneous recovery. Interestingly, this intervention impaired extinction. Finally, we demonstrated that this emotional remodeling to eliminate fear expression requires the activation of dopamine and oxytocin receptors during retrieval. Hence, these results shed new lights on the memory updating process and suggests that the exposure to rewarding information such as a female during retrieval fear memory in males reduces a previously consolidated fear memory.

Disclosures: A. Arellano Perez: None. **A. Kautzmann Sartori:** None. **S. Maren:** None. **L. de Oliveira Alvares:** None.

Poster

PSTR163. Neural Circuits of Fear and Aversive Processing

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR163.18/LL2

Topic: G.01. Fear and Aversive Learning and Memory

Support: R01MH065961

Title: Optogenetic inhibition of hippocampal-prefrontal projections facilitates fear extinction in rats

Authors: *S. L. PLAS, K. CRAYTON, S. MAREN;
Texas A&M Univ., College Station, TX

Abstract: Considerable work has revealed that the infralimbic cortex (IL) is critical for the extinction of conditioned fear in rats. The ventral hippocampus (VH) is a major source of excitatory input to the IL and has been implicated in extinction learning and retrieval. Our lab has demonstrated that VH projections to IL mediate the relapse of conditioned fear that occurs when an extinguished conditioned stimulus (CS) is encountered outside of the extinction context (i.e., “renewal"). Further, we have demonstrated that activation of VH→IL projections inside the extinction context impairs extinction retrieval. Electrophysiological recordings reveal that excitatory VH projections recruit IL inhibitory neurons to inhibit IL principal cell activity. Collectively, these data lead to the hypothesis that excitatory VH→IL projections not only oppose extinction retrieval, but also undermine extinction learning. To test this hypothesis, we used an optogenetic approach to inhibit VH terminals in the IL during extinction of auditory fear conditioning. To this end, AAV-CaMKII-ArchT-GFP, a light-activated outward proton pump, or AAV-CaMKII-GFP, was bilaterally infused into the VH of male and female rats. Seven weeks later, optic fibers were bilaterally implanted in the IL. After recovery, rats were submitted to auditory fear conditioning (context A; 5 tone-footshock trials), context B exposure, fear extinction with optogenetic manipulations (context B; 45 tone-alone trials), extinction retrieval (context B; 10 tone-alone trials), and renewal testing (context A; 45 tone-alone trials). Optogenetic inhibition of VH terminals in the IL did not affect conditioned freezing during the extinction session but led to a reliable reduction in freezing during both the CS and intertrial intervals throughout the subsequent retrieval test. Interestingly, during renewal testing in the conditioning context we observed a significant reduction in contextual freezing, as well as reduced renewal of freezing to the extinguished CS. These results suggest that VH→IL projections oppose extinction learning and promote the long-term expression of both contextual fear and fear to the CS.

Disclosures: S.L. Plas: None. **K. Crayton:** None. **S. Maren:** None.

Poster

PSTR163. Neural Circuits of Fear and Aversive Processing

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR163.19/LL3

Topic: G.01. Fear and Aversive Learning and Memory

Support: R01MH117852

Title: Brainstem-amygdala interactions lead to mPFC inhibition and impair extinction learning

Authors: *H. BAYER[1], A. BINETTE[1], J. HASSELL JR.[1], V. JULIANO[2], K. L. CRAYTON[1], S. MALIREDDY[1], C. D. MUNHOZ[2], S. MAREN[1];
[1]Texas A&M Univ., College Station, TX; [2]Univ. de Sao Paulo, Sao Paulo, Brazil

Abstract: The amygdala is a central node in the stress circuitry of the brain. It also encodes CS-US contingencies to modulate downstream targets and organize defensive behavior. Under stress, the amygdala suppresses the medial prefrontal cortex (mPFC), and this is thought to underlie extinction learning impairments. However, the neural circuits that activate the amygdala under stress are not well understood. It has been shown that activation of corticotropin releasing factor (CRF) neurons in the central amygdala (CeA) impairs extinction. Interestingly, CRF+ CeA neurons project to the locus coeruleus (LC), which projects to the basolateral amygdala (BLA). Here we explore the possibility that CeA CRF+ neurons drive an LC-BLA circuit to reduce mPFC activity and cause stress-induced extinction impairments. CRF-Cre transgenic rats received injections of either a blank virus or AAV-DIO-ChR2 and had optic fibers implanted above the CeA. One day after auditory fear conditioning, animals underwent fear extinction with optogenetic stimulation (20 sec, 10 Hz, 470 nm) during each CS presentation. Extinction retrieval was conducted 24 hrs later. During retrieval, the ChR2 group showed higher levels of freezing, suggesting that stimulation of CeA-CRH+ neurons impaired extinction learning. We next tested whether chemogenetic activation of LC-BLA projections would enable an immediate extinction deficit. We injected the BLA with a retrograde virus encoding an excitatory DREADD (CAV2-PRSx8-hM3Dq). Animals were injected with CNO or VEH before a weak fear conditioning session, received an extinction session 15 min after conditioning, and were tested for retrieval on the following day. The CNO treatment led to higher levels of freezing during both extinction and retrieval suggesting that LC-BLA activation induces an immediate extinction deficit. We then explored whether fear conditioning activates BLA-mPFC projections. Animals were injected with the retrograde tracer CTb in either the infralimbic (IL) or prelimbic (PL) cortices, and were perfused 2 hours after conditioning, context exposure, or staying in home-cage. BLA sections were stained for c-Fos and counted for CTb (+) and c-Fos (+) cells. In conditioned animals, BLA-IL projections were relatively more activated than BLA-PL, suggesting that BLA preferentially targets the IL under stress. Together, these results reveal a differential engagement of distinct BLA projections under stress and implicate upstream circuits that induce BLA activity in extinction learning impairments.

Disclosures: H. Bayer: None. **A. Binette:** None. **J. Hassell Jr.:** None. **V. Juliano:** None. **K.L. Crayton:** None. **S. Malireddy:** None. **C.D. Munhoz:** None. **S. Maren:** None.

Poster

PSTR163. Neural Circuits of Fear and Aversive Processing

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR163.20/LL4

Topic: G.01. Fear and Aversive Learning and Memory

Support: R01MH065961

Title: Neuronal activity in the thalamic nucleus reuniens during the conditioning and extinction of fear in male and female rats

Authors: *T. TUNA[1], M. S. TOTTY[2], S. PETERS[1], S. MAREN[1];
[1]Texas A&M Univ., College Station, TX; [2]TAMU, Lieber Inst. for Brain Develop., Baltimore, MD

Abstract: The nucleus reuniens (RE) is a midline thalamic structure that interconnects the medial prefrontal cortex and the hippocampus via bidirectional connections. We have recently identified a critical role for RE in the extinction of fear memory. This work suggests that RE neurons may have an active role in suppressing conditional fear responses, and single-unit recording data provide some support for this hypothesis. However, the learning-related responses of RE neurons during fear conditioning and extinction has not been performed. To address this, we recorded, in separate experiments, calcium transients and electrophysiological responses of RE neurons during both auditory fear conditioning, extinction, and extinction retrieval. In the fiber photometry experiment, adult male and female Long-Evans rats (n=8) were injected with AAV8-CaMKII-GCaMP6f and implanted with an optic fiber in RE. Relative to habituation, RE neurons exhibited robust US-evoked responses during conditioning and acquired CS-evoked responses by the end of the conditioning session. Contrary to our expectations, CS-evoked responses were maximal at the outset of extinction training, decreased over the course of several extinction trials, and remained low during extinction retrieval testing. Interestingly, spontaneous fluctuations in RE calcium activity were highly correlated with freezing behavior in an extinguished context: increases in RE activity reliably preceded transitions from freezing to activity. To ascertain the dynamics of individual RE neurons, we made single-unit recordings in the RE from adult male and female rats (n=13) during fear conditioning (n=41 cells) and extinction (n=33 cells). Fear conditioning was associated with robust CS- (n=12 of 41) and US-elicited (n=22 of 41) activity in RE neurons. During extinction, reliable CS-evoked responding was observed in roughly half of the neurons recorded. Roughly half of the responsive cells (n=7 of 16; “fear” neurons) showed maximal CS-evoked firing in the early extinction trials that decreased over the session. In contrast, the other population of cells (n=7; “extinction” neurons) showed the inverse pattern, firing to the CS only in the latest extinction trials. These data reveal that there is considerable heterogeneity in RE neuronal activity during extinction. “Extinction” neurons in the RE play a particularly important role in suppressing conditioned fear responses.

Disclosures: T. Tuna: None. **M.S. Totty:** None. **S. Peters:** None. **S. Maren:** None.

Poster

PSTR163. Neural Circuits of Fear and Aversive Processing

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR163.21/LL5

Topic: G.01. Fear and Aversive Learning and Memory

Support: R01MH117852

Title: Chemogenetic activation of the locus coeruleus mimics stress-impaired fear extinction in male and female rats.

Authors: *S. O. SWECK, S. MAREN, A. N. BINETTE;
Texas A&M Univ. Neurosci. Inst. For Neurosci., College Station, TX

Abstract: Extinction learning is an important mechanism for behavioral therapies of a variety of trauma- and stressor-related disorders. The infralimbic cortex (IL), a subdivision of the medial prefrontal cortex, has a critical role in fear extinction. Stress can induce impairments in fear extinction, including the "immediate extinction deficit" (IED) - an impairment that occurs when fear extinction is performed shortly after conditioning. Previous work in our lab suggests the IED is related to a stress-induced decrease in IL spike firing. Additionally, the IED can be induced or rescued by noradrenergic enhancement or blockade, respectively. Thus, we hypothesize that the locus coeruleus (LC), a major source of norepinephrine (NE) in the forebrain, mediates the stress-induced extinction deficit. To test this idea, we sought to describe the effects of chemogenetic activation of LC neurons on IL activity and behavior. Adult male and female (n=12) Long-Evans rats were bilaterally infused with AAV-PRSx8-hM3Dq-HA, an NE-specific excitatory designer receptor exclusively activated by designer drugs (DREADDs), in the LC and unilaterally infused with AAV-CaMKII-GCaMP6m into the IL. Additionally, a GRIN lens was implanted into the IL to allow for recording of calcium transients. To determine if LC-NE activation affects IL activity under basal conditions, animals underwent two days of recordings with injection of vehicle (VEH) or the DREADD ligand clozapine-N-oxide (CNO; 3 mg/kg, i.p.). We found that LC-NE activation drives increased freezing behavior and decreases IL activity by increasing the proportion of neurons suppressed after CNO injection relative to VEH. To ascertain whether LC-NE activation mimics footshock-induced changes in IL activity, animals were injected with VEH or CNO immediately prior to tone-only presentations or tone-footshock conditioning followed by 25 min of recording. Both footshock and LC-NE activation decreased IL principal activity with the former correlated to a weaker effect. Additionally, most recorded cells (67%) that were suppressed by LC-NE activation were also suppressed by footshock. Lastly, animals underwent standard delayed extinction and extinction retrieval protocols with either VEH or CNO onboard to determine if LC-NE activation prior to delayed extinction impairs retrieval and IL activity. CNO treatment prior to extinction resulted in higher levels of freezing during the first block of retrieval relative to VEH controls, suggesting that LC-NE activation is sufficient to impair fear extinction. Together, these results suggest that LC-NE signaling modulates IL activity and freezing behavior similarly to stress.

Disclosures: S.O. Sweck: None. **S. Maren:** None. **A.N. Binette:** None.

Poster

PSTR163. Neural Circuits of Fear and Aversive Processing

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR163.22/LL6

Topic: G.01. Fear and Aversive Learning and Memory

Support: R01MH065961

Title: Does the bed nucleus of the stria terminalis mediate circuit-induced relapse of extinguished fear?

Authors: *J. E. HASSELL, Jr., M. A. PARR, A. D. PEREZ, C. V. TERCILLA, S. MAREN; Texas A&M, College Station, TX

Abstract: Extinction learning is central to behavioral therapies for treatment of stressor- and trauma-related disorders including PTSD. We have recently discovered that the hippocampus (HPC) plays a critical role in the relapse of extinguished fear that occurs after pharmacological inactivation of the thalamic nucleus reuniens (RE) in rats. In particular, the HPC appears to encode contextual fear memories that are important for the "circuit-induced" relapse that occurs after RE inactivation. How these contextual memories drive increases in conditioned freezing behavior is unclear. However, there are abundant projections from the HPC to the bed nucleus of the stria terminalis (BNST) that may underlie relapse-induced freezing. Because the BNST has a prominent role in the expression of contextual fear memories, we hypothesize that it plays an important role in circuit-induced relapse. To test this idea, we explored whether pharmacological inactivation of the BNST would reduce the relapse of extinguished fear associated with RE inactivation. Adult male and female Long-Evans rats first underwent auditory fear conditioning followed twenty-four hours later by extinction. The next day, animals received intra-BNST infusions of either vehicle (saline) or the AMPA receptor antagonist, NBQX (10 ug/ul, 0.3 µl per side) (n = 6-8 per group) and intra-RE infusions of either vehicle (saline) or muscimol in a factorial design. Consistent with prior findings, RE inactivation caused a relapse of fear and increased freezing to the extinguished CS in the extinction context. However, preliminary data suggest that concurrent BNST inactivation does not attenuate this circuit-induced relapse. Further work will explore alternate neural pathways by which RE inactivation drives relapse-induced increases in freezing behavior.

Disclosures: J.E. Hassell: None. **M.A. Parr:** None. **A.D. Perez:** None. **C.V. Tercilla:** None. **S. Maren:** None.

Poster

PSTR163. Neural Circuits of Fear and Aversive Processing

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR163.23/LL7

Topic: G.01. Fear and Aversive Learning and Memory

Support: NIH Grant R01MH125916
NIH Grant F32MH119721

Title: Systems consolidation induces multiple memory engrams for a flexible recall strategy in observational fear memory in male mice

Authors: *J. TERRANOVA[1], J. YOKOSE[2], H. OSANAI[2], S. K. OGAWA[2], T. KITAMURA[3]; [1]Anat., Midwestern Univ., Downers Grove, IL; [2]Dept. of Psychiatry, [3]Dept. of Psychiatry and Neurosci., Univ. of Texas Southwestern Med. Ctr., Dallas, TX

Abstract: Although animals can learn to associate aversive stimuli and cues that predict danger from direct experience, this strategy is harmful and potentially lethal. Thus, strong evolutionary pressure favors vicarious learning in species ranging from zebrafishes to humans. Observational fear (OF) is the empathic response in which an observer witnesses and responds with fear to the demonstrator's aversive situation. In rodent models of OF, a fear conditioning apparatus is modified to contain an insulated observer chamber, a demonstrator chamber with an exposed shock grid floor, and a transparent partition separating both chambers. After a habituation period, an observer witnesses shocks delivered to the demonstrator and responds with fear. After OF conditioning, observers form a contextual fear memory of the context where they witnessed the demonstrator's aversive experience, a process called observational contextual fear conditioning (CFC). The anterior cingulate cortex (ACC) to amygdala neural circuit is essential for observers to acquire observational CFC memory. However, several outstanding questions regarding the behavioral and neurobiological aspects of Observational CFC memory remain. In this study, we expanded the behavioral model for recall of observational CFC memory and then examined the underlying neural circuit mechanisms in male mice. First, we showed that recall of observational CFC memory was long-lasting and, while observers could only recall observational CFC memory in the observer chamber at the recent (1 day) timepoint, they could recall observational CFC memory in both the observer and demonstrator chambers at the remote (28 day) timepoint. Recall in the demonstrator chamber was not fear generalization since observers did not exhibit elevated freezing levels even in a context that was similar to the demonstrator chamber. Next, we showed that recall of observational CFC memory at the recent timepoint was regulated by the dorsal hippocampus to BLA pathway, whereas recall of observational CFC memory at the remote timepoint in both chambers was regulated by the medial prefrontal cortex (mPFC) to BLA pathway. Finally, using chemogenetic neural silencing and activity-dependent cell labeling methods, we demonstrated that an observational CFC memory engram is rapidly generated in mPFC and, after systems consolidation, a new memory engram is generated and then activated when the observer recalls observational CFC memory in the demonstrator chamber at the remote timepoint. Our findings demonstrate the advantage of systems consolidation in a rodent model, allowing observers to make new inferences about a previously encountered situation.

Disclosures: J. Terranova: None. **J. Yokose:** None. **H. Osanai:** None. **S.K. Ogawa:** None. **T. Kitamura:** None.

Poster

PSTR163. Neural Circuits of Fear and Aversive Processing

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR163.24/LL8

Topic: G.01. Fear and Aversive Learning and Memory

Support: NIH Grant R21MH132052
NIH Grant KL2TR002245

Title: Nucleus accumbens core single cell ensembles bidirectionally respond to experienced versus observed aversive stimuli

Authors: O. DINCKOL[1], J. ZACHRY[2], ***M. G. KUTLU**[1];
[1]Cell Biol. and Neurosci., Rowan-Virtua Sch. of Osteo. Med., Stratford, NJ; [2]Vanderbilt Univ., Nashville, TN

Abstract: Empathy is the ability of adopting others' sensory and emotional states and is an evolutionary conserved trait among mammalians. In rodents, empathy manifests itself as social modulation of aversive stimuli such as acknowledging and acting on conspecifics' distress. The neuronal network underlying social transmission of information is known to overlap with the brain regions that mediate behavioral responses to aversive and rewarding stimuli. In this study, we aimed to identify single cell ensembles within the nucleus accumbens (NAc) core that respond to experienced and/or observed aversive stimuli using in vivo optical imaging of calcium activity via miniature scopes. Our results showed that experienced and observed aversive stimuli evoke NAc core ensemble activity that is largely positive, with a smaller subset of negative responses. The size of the NAc single cell ensemble response was greater for experienced aversive stimuli as compared to observed aversive events. Our results also revealed a subpopulation within the NAc core single cell ensembles that show a bidirectional response to experienced versus observed aversive stimuli (i.e., negative response to experienced and positive response to observed aversive stimuli). These results suggest that the NAc plays a role in differentiating somatosensory experience from social observation of aversion at a single cell level. This has important implications for psychopathologies where social information processing is maladaptive, such as autism spectrum disorders.

Disclosures: O. Dinckol: None. **J. Zachry:** None. **M.G. Kutlu:** None.

Poster

PSTR164. Neural Circuits of Fear and Aversive Processing: Amygdala Circuits

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR164.01/LL9

Topic: G.01. Fear and Aversive Learning and Memory

Support: NIH Grant R01 MH066958
NIH Grant R01 MH119283
Southeast Louisiana VA Merit grant I01 BX005118

Title: Noradrenergic alpha1 and beta receptors oppositely impact BLA pyramidal neuron excitability

Authors: *P. S. TIRRELL[1,2], X. FU[1,2], J. G. TASKER[1,3];
[2]Neurosci. Program, [3]Cell and Mol. Biol., [1]Tulane Univ., New Orleans, LA

Abstract: Regulating the balance of excitation and inhibition in the amygdala is imperative for adaptive emotional and behavioral responses to stressful stimuli and neuron recruitment into discrete fear memory traces. To help orchestrate complex neural processing and construct optimal context-specific fear memories, stress-induced release of norepinephrine (NE) from locus coeruleus (LC) terminals in the basolateral amygdala (BLA) activates a diverse array of adrenergic receptors that are expressed on distinct synapses, cellular compartments, and neuron types including Gq-coupled α1 adrenoceptors and Gs-coupled β adrenoceptors, both of which have been implicated in neuropsychiatric disorders. NE facilitates acute increases in BLA excitability by directly potentiating glutamatergic pyramidal neurons and dynamically altering interneuron activity. However, NE has both excitatory and inhibitory effects on pyramidal neurons in the BLA, suggesting NE's role in pyramidal neuron excitability is far more complex. To determine the relative contributions of different adrenoreceptor subtypes to BLA pyramidal neuron excitability, whole-cell patch clamp recordings were performed in brain slices from C57BL/6 wildtype mice and ARα1A and ARα1B knockout mice. In the presence of propranolol to block β adrenoceptors, norepinephrine (10 uM) activation of α1 adrenoceptors induced a tonic outward current in the presence of β adrenoceptor blocker propranolol. This α1-mediated tonic outward current hyperpolarized the membrane potential and blocked action potentials and was mimicked with the α1-specific agonist phenylephrine. The reversal potential of the outward current determined by voltage ramps revealed a potassium current likely mediated by activation of G-protein coupled inwardly rectifying potassium (GIRK) channels. NE activation of β receptors in the presence of the α1 antagonist prazosin caused a tonic inward current that depolarized the resting membrane potential and increased action potentials. These preliminary data suggest that BLA pyramidal neurons with greater β than α1 adrenoreceptor activation could bias a cell toward a higher state of excitability and make it susceptible to recruitment into a discrete fear memory engram.

Disclosures: P.S. Tirrell: None. **X. Fu:** None. **J.G. Tasker:** None.

Poster

PSTR164. Neural Circuits of Fear and Aversive Processing: Amygdala Circuits

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR164.02/LL10

Topic: G.01. Fear and Aversive Learning and Memory

Support: Southeast Louisiana VA Merit grant I01 BX005118

Title: Gq activation in cholecystokinin-positive interneurons in the basolateral amygdala ablates fear memory recall in adolescent mice

Authors: *M. T. WATSON[1], J. G. TASKER[2];
[1]Tulane Brain Inst., [2]Cell and Mol. Biol., Tulane Univ., New Orleans, LA

Abstract: Hyperactivity of the amygdala, a key region for adaptive stress and fear processing, is observed in Posttraumatic Stress Disorder (PTSD), which affects approximately 8% of the general population. This hyperactivity is suggested to be the cause of hyperarousal symptoms of PTSD, which include avoidance of cues related to the trauma, hypervigilance, and increased startle response. The loss of inhibitory tone within the basolateral nucleus of the amygdala (BLA) can result in long-term maladaptive stress processing. Perisomatic inhibitory interneurons, or basket cells, consist of cholecystokinin-positive (CCK) and parvalbumin-positive (PV) interneurons, which strongly regulate principal neuron output. Using a chemogenetic approach, we sought to characterize the respective roles of CCK and PV basket cells in associative fear learning, focusing on fear memory recall and extinction, and to determine whether CCK and/or PV neuron perisomatic inhibition is altered to generate hyperarousal symptoms following traumatic stress. Four- to 5-week-old male and female CCK-ires-Cre and PV-Cre mice on a C57BL/6J background received bilateral injections in the BLA of adeno-associated virus (AAV) vectors expressing Cre-dependent Gq-DREADD driven by a GABA neuron-specific promoter (AAV9-hDLX-DIO-hM3D-mCherry). Following recovery, mice were subjected to a 4-day fear conditioning protocol consisting of Day 1 (Acquisition): 7 auditory tones (CS) paired with foot shocks (US, 0.7 mA, 2 s); Day 2 (Recall/Extinction): 30 unpaired CS; Day 3 (Extinction): 30 unpaired CS; Day 4 (Extinction Recall): 15 unpaired CS. Clozapine N-oxide (CNO) was administered IP to drive activation of the Gq-coupled DREADD 30 min prior to Extinction trials. Gq-DREADD activation in BLA CCK neurons resulted in a robust reduction of freezing behavior compared to controls; the freezing behavior recovered to control levels on Day 4 Extinction Recall, suggesting that Gq-DREADD activation in CCK neurons abolished fear memory recall, but not the fear memory or its extinction. Gq-DREADD activation in PV neurons failed to block the fear recall, suggesting a different role for PV neurons in fear memory formation. Neither response was affected by traumatic stress from a traumatic exposure with reminders paradigm. These results suggest that BLA CCK and PV basket cells differentially modulate fear memory recall, and that this does not appear to be impacted by traumatic stress. Continued circuit and cellular-level characterization of perisomatic inhibition in the BLA during associative fear conditioning could lead to better therapeutic strategies for patients suffering from PTSD

Disclosures: M.T. Watson: None. **J.G. Tasker:** None.

Poster

PSTR164. Neural Circuits of Fear and Aversive Processing: Amygdala Circuits

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR164.03/LL11

Topic: G.01. Fear and Aversive Learning and Memory

Support: Southeast Louisiana VA Merit grant I01 BX005118

Title: Traumatic Stress Disruption of Basolateral Amygdala Inhibitory Circuits

Authors: *B. L. W. SWEETEN, M. T. WATSON, X. FU, J. G. TASKER;
Tulane Univ., New Orleans, LA

Abstract: Traumatic stress is thought to disrupt the excitatory and inhibitory balance within the basolateral nucleus of the amygdala (BLA), an area responsible for much of the synaptic plasticity that occurs during and following stress and emotional arousal. Norepinephrine (NE) in the BLA plays a critical role in emotional arousal. Parvalbumin-expressing (PV) GABAergic interneurons in BLA are important for gating principal neuron excitability. Recently, we showed that NE activation of Gq-coupled α1 adrenoreceptors or Gq-DREADD activation in PV interneurons elicits a non-canonical, highly stereotyped synchronized bursting pattern of inhibitory postsynaptic currents (IPSCs) in BLA principal neurons that facilitates fear memory recall by suppressing gamma oscillations (Fu et al., 2022). Here, we assessed how exposure to a traumatic stress paradigm causes circuit changes and behavioral state changes characteristic of post-traumatic stress disorder (PTSD). Four- to six-week-old male and female PV-Cre or wild type littermates were either bilaterally injected with AVV-hdlx-Gq-DREADD in BLA or left as controls. Following a 1-week recovery, mice were exposed to a modified Traumatic Experience with Reminders of Stress (TERS) paradigm that consisted of a 5-min exposure to a pre-shock context followed by a 10-s, 2 mA 'traumatic' foot shock in a second context. Subsequently, the mice were reminded of the traumatic shock experience by exposure to the pre-shock context for 1 min once per day for the next 5 days. The mice were tested for their acoustic startle response (ASR: 15, 110 dB white noise bursts, 30s ITI) one day prior to TERS exposure and retested on the day following the TERS exposure. A subset of mice were subjected to a 4-day fear conditioning protocol that began 5 days later, followed by a final ASR test. Whole-cell patch clamp recordings of NE and Gq modulation of inhibitory synaptic activity were performed in principal neurons in amygdala slices 7 to 10 days following the TERS in both DREADD-expressing and control mice. TERS-exposed mice had significantly higher ASR ($p<0.001$) compared to their baseline and showed greater fear memory acquisition ($p<0.01$) and a delayed fear extinction ($p<0.001$) compared to control mice. Principal neurons recorded from TERS-exposed mice displayed a loss of the PV neuron-mediated stereotypical bursting pattern of IPSCs in response to NE (100 μM) and CNO (5 μM). These results indicate that the modified TERS paradigm generates a behavioral state characteristic of PTSD in a mouse model, and that this may be mediated by altered patterns of activity in BLA PV neuron inhibitory circuits.

Disclosures: B.L.W. Sweeten: None. **M.T. Watson:** None. **X. Fu:** None. **J.G. Tasker:** None.

Poster

PSTR164. Neural Circuits of Fear and Aversive Processing: Amygdala Circuits

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR164.04/LL12

Topic: G.01. Fear and Aversive Learning and Memory

Title: Parallel processing of noxious stimuli in the basolateral amygdala circuits

Authors: *G. NAGY[1,2], D. MAGYAR[3,2], J. VERES[1], C. PARDO-BELLVER[4], Z. REÉB[1,5], B. BARABAS[3,2], P. FOLDI[1,2], N. HAJOS[3];
[1]Inst. of Exptl. Med., Budapest, Hungary; [2]Semmelweis Univ., Budapest, Hungary; [3]Indiana Univ., Bloomington, IN; [4]Univ. of Valencia, Valencia, Spain; [5]Eotvos Lorand Univ., Budapest, Hungary

Abstract: The basolateral amygdala (BLA) is composed of distinct nuclei, including the lateral (LA) and basal nuclei (BA). The BLA plays a critical role in Pavlovian fear conditioning. In this robust learning paradigm, a neutral conditioned stimulus becomes associated to a biologically relevant unconditioned stimulus (US). Noxious stimuli, such as mild electrical shocks, proved to be a highly potent US and thus are widely applied in studies of fear learning. Therefore, here we aim to precisely describe the elements of the BLA neural network that participate in the coding of the noxious stimuli. For this, in awake head-fixed and anesthetized mice we used silicon probes to simultaneously record single-unit activity in the LA and BA during the presentation of mild electrical shocks. In addition, in anesthetized mice we juxtacellularly recorded and labeled randomly sampled neurons in the LA and BA during the administration of electrical shocks. Our data revealed that two populations of responsive principal cells (PCs) could be distinguished based on the latency of their stimulus-evoked firing. In the first population, PCs responded with a short latency to the noxious stimuli, while the stimulus related elevation of firing rates in the second group occurred with a longer latency. Both cell groups were present in both LA and BA. In further experiments we described the latency of the noxious stimulus-evoked responses of two subpopulations of BA neurons projecting to the dorsomedial striatum (DMS) or to the medial prefrontal cortex (mPFC). For this, we expressed channelrhodopsin-2 in these neurons in a projection-specific manner using retrograde adeno-associated viruses. We used silicon probes to record the stimulus evoked responses of the optotagged DMS-projecting or mPFC-projecting BA neurons. Our results show that these subpopulations show markedly distinct responses to noxious stimulation. Together, these results indicate that the LA and BA process noxious inputs simultaneously and that the shock-evoked response latencies in the BA occur in a projection-specific manner.

Disclosures: G. Nagy: None. **D. Magyar:** None. **J. Veres:** None. **C. Pardo-Bellver:** None. **Z. Reéb:** None. **B. Barabas:** None. **P. Foldi:** None. **N. Hajos:** None.

Poster

PSTR164. Neural Circuits of Fear and Aversive Processing: Amygdala Circuits

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR164.05/Web Only

Topic: G.01. Fear and Aversive Learning and Memory

Support: NSERC
CIHR

Title: Connectivity between basolateral amygdala and olfactory and auditory cortices in second-order threat conditioning: A retrograde tracing study in the rat

Authors: *T. SEPAHVAND, K. D. POWER, A. M. JANES, Q. YUAN;
Mem. Univ. of Newfoundland, St. John's, NL, Canada

Abstract: In humans, fear memories—which are involved in conditions such as post-traumatic stress disorder (PTSD)—are highly complex and may be triggered by stimuli or events not directly related to the initial traumatic experience. Animal models using paradigms such as Pavlovian second-order threat conditioning (SOC), however limited in their ability to recapitulate human experiences, attempt to capture some of this complexity. Here, we aimed to explore the mechanisms underlying the formation of higher-order threat memories at a systems and cellular level by using retrograde tracing (cholera toxin subunit B (CTB)) in brain areas of interest including the basolateral amygdala (BLA), a key convergence site for sensory and affective information, and auditory and olfactory (piriform) sensory cortices. Adult (3-6 months) Sprague Dawley rats of both sexes were trained to associate a tone (CS1) with a shock (US) to produce first-order conditioning, then an odor (CS2) with the previous CS1 to create SOC. A CS1/CS2 unpaired group was used as a control. Rats received intra-BLA, -piriform, and -auditory cortex infusions of fluorophore-conjugated CTB prior to SOC. cFos immunohistochemistry (IHC) and fluorescence microscopy were used to assess CTB labeling and co-localization with cFos in areas of interest. Sections from CTB brains are currently undergoing fluorescence imaging and analysis to visualize and compare the precise afferent and efferent subpopulations of BLA and piriform and auditory cortices involved in SOC. Our preliminary results highlight the role of reciprocal connections between the BLA and auditory cortex in SOC memory formation, implicating the role of the auditory cortex in SOC memory formation. So far, our results are in line with a more recent conceptualization of sensory cortices as more than mere sensory processors. An optogenetic activation experiment will be used to assess the functional significance of these specific subpopulations in SOC. In conclusion, sensory cortices and the amygdala work together to store SOC threat memories, which has implications for the treatment of complex fear memories such as those in PTSD.

Disclosures: T. Sepahvand: None. **K.D. Power:** None. **A.M. Janes:** None. **Q. Yuan:** None.

Poster

PSTR164. Neural Circuits of Fear and Aversive Processing: Amygdala Circuits

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR164.06/LL13

Topic: G.01. Fear and Aversive Learning and Memory

Title: Roles of the central amygdala to ventral tegmental area projections in fear regulation and the mechanism study

Authors: *C. LI, X.-M. LI;
Zhejiang Univ., Hangzhou, China

Abstract: Fear is essential to an individual's survival. Abnormal fear will make people or animals more vulnerable to danger, and even lead to a series of neuropsychiatric diseases. Previous studies have suggested that functional abnormalities of the ventral tegmental area (VTA) may lead to fear-related neuropsychiatric diseases, such as post-traumatic stress disorder and schizophrenia, but the underlying mechanisms remain unclear. Our preliminary data has firstly shown that the projection from the central amygdala (CeA) to the ventral tegmental area could regulate fear. However, the specific cell types and molecular mechanisms of fear regulation remain elusive. We will study the role and mechanism of the CeA-VTA circuit in fear emotion from multiple hierarchical systems including cell types, neural circuits, and molecular mechanisms by combining electrophysiology, in vivo optical fiber recording, animal behavior, and molecular genetics. The results are expected to further reveal the neural mechanism of fear and provide new ideas and potential therapeutic targets for the treatment of fear and related diseases.

Disclosures: C. Li: None.

Poster

PSTR164. Neural Circuits of Fear and Aversive Processing: Amygdala Circuits

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR164.07/LL14

Topic: G.01. Fear and Aversive Learning and Memory

Support: Max-Planck Society
European Research Council Grant (no. 885192, BrainRedesign)

Title: NDNF GABAergic neurons regulate aversive stimuli in the basolateral amygdala

Authors: *Y. MASTRODICASA, R. KLEIN;
Max Planck Inst. for Biol. Intelligence, Munich - Planegg, Germany

Abstract: The basolateral amygdala (BLA) is an integration center that regulates responses to both positive and negative behavioral stimuli. Primarily formed by excitatory pyramidal cells, the role that local inhibitory microcircuits play in regulating BLA activity and in mediating amygdala-related cognitive processes is not well understood. Our study provides evidence for a role of neuron-derived neurotrophic factor (NDNF)-positive neurons in the regulation of neural activity in the BLA and for their responsiveness to negative emotional stimuli. Using the

expression of the immediate early gene c-Fos as a marker for NDNF activation in the BLA, we found that when animals were tested during fear conditioning and odor-induced innate fear paradigms, the percentage of NDNF-c-Fos-positive cells in the BLA significantly increased compared to control conditions. Moreover, chemogenetic and optogenetic loss-of-function experiments confirmed that NDNF neurons are in fact necessary to regulate BLA responses to aversive stimuli during fear acquisition and odor-induced innate fear. Furthermore, we employed *ex vivo* patch clamp recordings and monosynaptic rabies tracing studies to identify the circuits that NDNF neurons form both within the BLA and with other brain regions. Our tracing studies identified significant inputs from the cortical amygdala area (CoA) to the BLA-NDNF neurons which might play an important role in the regulation of innate behaviors and responses to threatening stimuli. Further exploration will be necessary to understand how NDNF GABAergic neurons contribute to the modulation of BLA neuronal circuits and their overall response to negative states, thus contributing to animal behavior.

Disclosures: Y. Mastrodicasa: None. **R. Klein:** None.

Poster

PSTR164. Neural Circuits of Fear and Aversive Processing: Amygdala Circuits

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR164.08/LL15

Topic: G.01. Fear and Aversive Learning and Memory

Support: National Natural Science Foundation of China (82090030, 32071022, 32100815 and 82288101)
STI2030-Major Projects (2021ZD0202700)
Key-Area Research and Development Program of Guangdong Province (2018B030334001 and 2019B030335001)
Key R&D Program of Zhejiang Province (2020C03009 and 2021C03001)
Fundamental Research Funds for the Central Universities (2021FZZX001-37)
CAMS Innovation Fund for Medical Sciences (2019-I2M-5-057)
Innovative and Entrepreneur Team of Zhejiang for 2020 Biomarker-Driven Basic and Translational Research on Major Brain Diseases (2020R01001) and the fellowship of China Postdoctoral Science Foundation (2020M681833)

Title: Distinct Circuits from Central Lateral Amygdala to Ventral Part of Bed Nucleus of Stria Terminalis Regulate Different Fear Memory

Authors: Y. ZHU[1,2], ***S. XIE**[1], A. PENG[1], X. YU[1], C. LI[1,2], X. LI[1,2,3];
[1]Zhejiang Univ., Hangzhou, China; [2]Zhejiang Univ. Sch. of Med., Hangzhou, China; [3]Chinese Acad. of Med. Sciences/Guangdong–Hong Kong–Macao Greater Bay Area Ctr. for Brain Sci. and Brain-Inspired Intelligence, Guangzhou, China

Abstract: The ability to differentiate stimuli predicting fear is critical for survival, however, the relevant molecular and circuit mechanism remain poorly understood. Using *in vivo* transsynaptic circuit-dissecting anatomical approaches, we identify the projections from central lateral amygdala (CeL) protein kinase C δ (PKCδ) positive neurons and somatostatin (SST) positive neurons to the ventral part of bed nucleus of stria terminalis (vBNST) GABAergic and glutamatergic neurons. Manipulation of $PKC\delta^{CeL\text{-}vBNST}$ pathway specifically regulated context fear memory, whereas $SST^{CeL\text{-}vBNST}$ pathway regulated tone fear memory. Further, our optogenetic studies demonstrate either prolonged-activation or inhibition of the circuits reduced fear memory. Interestingly, optogenetic manipulation of vBNST neurons received the projection from $PKC\delta^{CeL}$ or SST^{CeL} neurons could bidirectionally regulate context or tone fear memory respectively. The paradoxical behavioral result of projection-manipulation versus downstream-neuron-manipulation indicated the possible mechanism on the pre-synapse of the circuits. We then proved the existence of δ and κ opioid receptor protein expression in the CeL-vBNST circuits. Administration of an opioid receptor antagonist cocktail on the $PKC\delta^{CeL\text{-}vBNST}$ or $SST^{CeL\text{-}vBNST}$ pathway rescued context or tone fear reduction induced by prolonged activation of the circuits. Together, these findings establish a functional role for distinct CeL-vBNST circuits in differently regulating and properly maintaining fear.

Disclosures: **Y. Zhu:** None. **S. Xie:** None. **A. Peng:** None. **X. Yu:** None. **C. Li:** None. **X. Li:** None.

Poster

PSTR164. Neural Circuits of Fear and Aversive Processing: Amygdala Circuits

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR164.09/LL16

Topic: G.01. Fear and Aversive Learning and Memory

Title: Excitatory synaptic inputs on parvalbumin expressing and axo-axonic cells are altered distinctly upon associative fear learning and aversive stimuli in the basal amygdala.

Authors: J. M. VERES[1], Z. FEKETE[1,2], ***T. ANDRASI**[3,1], L. ROVIRA-ESTEBAN[1], O. I. PAPP[1], B. BARABAS[3,1,2], N. HAJOS[3,1];
[1]Lab. of Network Neurophysiol., ELRN Inst. of Exptl. Med., Budapest, Hungary; [2]Szentagothai Doctoral Sch. of Neurosciences, Semmelweis Univ., Budapest, Hungary; [3]Psychological and Brain Sci., Indiana Univ., Bloomington, IN

Abstract: In cortical structures, parvalbumin-containing perisomatic inhibitory neurons, namely basket cells (PVBCs) and axo-axonic cells (AACs), are in a position to effectively control local excitatory neuronal activity during cognitive processes, such as learning. In order to uncover the role of these inhibitory neuron types in aversive learning, we investigated the effect of associative fear conditioning and aversive stimuli on excitatory neurotransmission received by PVBCs and AACs in the basal amygdala (BA), which is a known locus of fear related learning in the mammalian brain. To reveal the features of excitatory synaptic inputs on BA PVBCs and

AACs, we combined a fear conditioning learning paradigm in BAC-PV-eGFP mice immediately followed by *in vitro* electrophysiological recordings. We compared the properties of miniature excitatory postsynaptic currents (mEPSCs) in three groups of mice: in group 1) mice received tone presentations only (conditioned stimulus, CS group); in group 2) mice were presented a tone and a mild electrical foot shock unpaired 7 times (unsigned unconditioned stimulus, unsigned US group); and in group 3) mice received a tone co-terminated with a mild electrical foot shock 7 times (signed US group). Our results revealed that the peak amplitude and rate of mEPSCs in PVBCs decreased significantly in groups that received US, either signed or unsigned, compared to those mice in CS group. Furthermore, mice in the unsigned US group showed a significant decrease in both parameters compared to those mice in the signed US group. In contrast, in AACs the rate of mEPSCs was increased significantly in both unsigned US and signed US groups in comparison to recordings accomplished in the CS group. However only the unsigned US group showed a modest decrease in the peak amplitude compared to the CS and signed US group. These results suggest that the cue-related fear learning and unsigned aversive stimuli differently alter excitatory synaptic inputs in PVBCs and AACs in the BA. Our results support the hypothesis that these perisomatic inhibitory cell types play different roles in aversive signal processing depending on BA function.

Disclosures: **J. M. Veres:** None. **Z. Fekete:** None. **T. Andrasi:** None. **L. Rovira-Esteban:** None. **O.I. Papp:** None. **B. Barabas:** None. **N. Hajos:** None.

Poster

PSTR164. Neural Circuits of Fear and Aversive Processing: Amygdala Circuits

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR164.10/LL17

Topic: G.01. Fear and Aversive Learning and Memory

Support: Foundation For OCD Research
Berkelhammer Award for Excellence in Neuroscience

Title: Control of negative reinforcement and extinction by intercalated nuclei of the amygdala

Authors: *R. ST. LAURENT[1], K. M. KUSCHE[3], A. C. KREITZER[3,4,5], R. MALENKA[2]; [1]Psychiatry and Behavioral Sci., [2]Nancy Pritzker Laboratory, Dept. of Psychiatry and Behavioral Sci., Stanford Univ., Stanford, CA; [3]Gladstone Inst., San Francisco, CA; [4]MapLight Therapeut., Redwood City, CA; [5]Depts of Physiol. and Neurol., UCSF, San Francisco, CA

Abstract: Front-line treatments for obsessive compulsive disorder (OCD) are successful only in a fraction of the patient population. Thus there is a need for a high-resolution understanding of circuit dysfunction that drives symptoms for the advancement of more efficacious therapeutic interventions. One subregion of the amygdala, the intercalated nuclei (ITC), likely contributes to OCD circuit dysfunction. However, further work is needed to identify specific maladaptive ITC circuits. The ITC is comprised of clusters of inhibitory cells that mediate behavior during classic

fear learning and extinction, a form of reinforcement learning. Aberrant gating of neural activity at this interface can disrupt normal feedforward inhibitory circuits, leading to disrupted negative reinforcement behavior. However, whether the same ITC circuits regulate negative reinforcement within a *dynamic* environment, such as approach-avoidance behavior, remains unknown.
We used adult male and female FoxP2-IRES-cre mice in behavioral procedures including platform-mediated avoidance, open field, elevated plus maze, von Frey, and real-time place preference. Cre-dependent GCamp6f, ChR2, eNphR3.0, or mCherry were injected bilaterally into the dorsal ITC cluster 4-8 weeks prior to experiments. In a subset of experiments, we crossed FoxP2-IRES-cre mice with SAPAP3-null mouse strain to generate wildtype and knockout littermates.
In fiber photometry recordings of ITC calcium activity in mice, ITC cells within the dorsal cluster were activated by both positive and negative stimuli in an approach-avoidance task. We next activated or silenced the dorsal ITC using channelrhodopsin or halorhodopsin, respectively, during extinction of the warning tone that predicts a footshock in the approach-avoidance task. Optogenetic stimulation increased avoidance and silencing enhanced the extinction of avoidance. We also performed photometry recordings in SAPAP3 knockout mice, a commonly used model for OCD, and found diminished responses to footshock compared to wildtype littermates. SAPAP3 knockout mice also exhibited higher baseline avoidance behaviors compared to wildtype littermates. We photo-stimulated the dorsal ITC as in the prior experiments and found that avoidance behavior was similarly increased by activation and reduced by silencing dorsal ITC activity. Together, these results provide information about the temporal dynamics of dorsal ITC neuronal activity during approach-avoidance behavior. Furthermore, we demonstrate a causal role for dorsal ITC activity during extinction of negative reinforcement in both wildtype and SAPAP3 knockout mice.

Disclosures: **R. St. Laurent:** None. **K.M. Kusche:** None. **A.C. Kreitzer:** A. Employment/Salary (full or part-time); MapLight Therapeutics. **R. Malenka:** None.

Poster

PSTR164. Neural Circuits of Fear and Aversive Processing: Amygdala Circuits

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR164.11/LL18

Topic: G.01. Fear and Aversive Learning and Memory

Support: R01 MH122561

Title: Role of divergent central amygdala projections in defensive response regulation in mice

Authors: *C. BORKAR[1], C. E. STELLY[2], Q.-S. E. LE[1], C. FRIEDMAN[1], J. P. FADOK[3]; [2]Cell & Mol. Biol., [3]Psychology and Tulane Brain Inst., [1]Tulane Univ., New Orleans, LA

Abstract: Organisms exhibit a repertoire of adaptive responses to survive threats. However, traumatic events impact psychological well-being, which can lead to hyperactive responses to stimuli, responses which are observed in mental illnesses like generalized anxiety, post-traumatic stress and panic disorders. Our previous studies show that the neuronal circuits in the central nucleus of the amygdala (CEA) are vital for regulating adaptive defensive freezing and flight. Moreover, prefrontal inputs to CEA affect the selection of flight responses in mice. However, downstream pathways of CEA that regulate high intensity defensive responses are unknown. The periaqueductal gray (PAG) is a well-known target nucleus of CEA for fear regulation, and the retrorubral field (RRF) is a mid-brain structure recently observed to regulate threat associated responses, raising the possibility that direct projections of CEA to PAG and RRF could modulate fear response switching. Here, we established the neuroanatomical connections between CEA-PAG and CEA-RRF using anterograde and retrograde tracings. We found that CEA sends a strong projection to both of these regions. We also noted that CEA corticotrophin releasing hormone and somatostatin positive neurons innervate dopamine neurons in RRF. Functional connectivity was established by ablating CEA neuronal inputs to these regions using viral vectors expressing caspase. The caspase vector injected mice were then subjected to a modified Pavlovian fear conditioning paradigm that elicits freezing and flight responses to distinct components of a serial compound stimulus. We show that deleting CEAàRRF projecting neurons significantly reduced both freezing and flight responses. Deleting the PAG neurons that are innervated by the CEA also significantly affects freezing behavior. Electrophysiological data suggests that prefrontal neurons innervate PAG- and RRF-projecting CEA neurons. We are currently undertaking optogenetic and chemogenetic manipulations to demonstrate their sufficiency for defensive response scaling. Collectively, our results demonstrate that the CEA exerts direct top-down control over PAG and RRF neuronal circuits to execute defensive fear scaling.

Disclosures: C. Borkar: None. **C.E. Stelly:** None. **Q.E. Le:** None. **C. Friedman:** None. **J.P. Fadok:** None.

Poster

PSTR164. Neural Circuits of Fear and Aversive Processing: Amygdala Circuits

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR164.12

Topic: G.01. Fear and Aversive Learning and Memory

Support: NIH MH122023
NSF OAC-1730655

Title: Information processing in a model lateral amygdala network

Authors: *G. GLICKERT[1], B. LATIMER[2], P. SAH[4], S. S. NAIR[3];
[1]Univ. of Missouri, Columbia, Columbia, MO; [3]Electrical & Computer Engin., [2]Univ. of Missouri, Columbia, MO; [4]The Univ. of Queensland Queensland Brain Inst., St Lucia, Australia

Abstract: In a cued Pavlovian fear conditioning, rodents were exposed to repeated pairing of a conditioned stimulus (CS) tone with an aversive unconditioned stimulus (US) footshock. This paradigm has been shown to lead to enhancement (LTP) of synapses carrying CS information to the lateral amygdala (LA) by a process that requires N-methyl-D-aspartate receptors (NMDAR). However, the exact role of NMDARs is not understood. Here we use a 5000-cell computational model of the lateral amygdala (LA) that consisted of principal neurons and fast spiking interneurons (FSIs), to explore the role of NMDA receptors in signal transmission during CS tones. The model is constrained using biological data, including single unit multi-electrode data (up to 30 cells, at 25 kHz), in runs with- and without- blockade of NMDA receptors.
We first considered the habituation phase with CS tones presented alone. After 12 tone trials the pyramidal cell response to the CS decreased from 1.3 to 1 Hz due to depotentiation of tone-pyramidal synapses, which the model predicted as the underlying mechanism for habituation. Next we examined the tone response with, and without NMDAR-block. Biological data showed that the firing of pyramidal cells remained relatively unchanged while the FS neuron firing rates decreased considerably. The model similarly had a relatively unchanged firing rate of ~0.9 Hz in both conditions, while there was a 42% decrease in activity from 27±7 to 11.7±3 Hz for the interneurons. The model predicts that this change results from a decrease in NMDAR activity at CS synapses to principal neurons, and FS interneurons as well as reduction in principal neuron to principal neuron synapses. Interestingly, the reduction in excitation to principal cells was compensated by a concomitant reduction in inhibition due to the reduced FSI firing, thus principal cell firing rates remained unchanged. The reduction in the firing rate of FSIs with the block was found to result from a larger role for NMDAR transmission via the excitatory tone synapses onto FSIs. Further supporting this observation, the model showed that FSIs with high firing rates had significantly higher NMDA currents (averaged over 50 ms prior to spike) compared to those with low firing rates ($p < 0.01$). Taken together, these preliminary findings suggest an important role for NMDARs in information processing in the amygdala, particularly via the recruitment of interneurons.

Disclosures: **G. Glickert:** None. **B. Latimer:** None. **P. Sah:** None. **S.S. Nair:** None.

Poster

PSTR165. Motivation and Food

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR165.01/LL19

Topic: G.03. Motivation

Support: NIH 5R01DA015188
NIH 5R01MH063649
NIH 1F31MH125613
NIH 1F99NS124176

Title: Optogenetic hedonic hotspots in orbitofrontal cortex, insula, ventral pallidum, and anterior cingulate cortex causally enhance liking and wanting for sweet food reward

Authors: *I. MORALES[1], J. KRAVCHENKO[1], K. E. EMERY[2], M. AYOUB[1], K. R. URSTADT[3], D. C. CASTRO[4], K. C. BERRIDGE, PHD[2];
[2]Dept. of Psychology, [1]Univ. of Michigan, Ann Arbor, MI; [3]Dept. of Cognitive Sci., Occidental Col., Los Angeles, CA; [4]Sch. of Med. Mallinckrodt Inst. of Radiology, Washington Univ. in St. Louis, St. Louis, MO

Abstract: Reward consists of learning, 'wanting', and 'liking' components, but 'liking' remains the least understood. 'Liking', or hedonic impact can be causally amplified by opioid and orexin stimulation in brain hedonic hotspots of the nucleus accumbens, ventral pallidum, orbitofrontal cortex, and insula cortex. 'Liking' enhancement can be measured as increases in the number of positive hedonic orofacial expressions elicited by oral infusions of sucrose solutions in the affective taste reactivity test. A question arises of whether hedonic hotspots are mere neurochemical artifacts of local opioid/orexin drug microinjections or more robust hedonic entities that can also be revealed by other means. Here we present novel data showing that optogenetic stimulation in hedonic hotspots of the insula, OFC, and ventral pallidum doubles the number of hedonic 'liking' reactions elicited by sweet taste. Hedonic enhancement was anatomically restricted to the hedonic hotspots with the same sites as borders as previously identified by drug microinjections. Outside of the hedonic hotspots optogenetic stimulation failed to increase 'liking', but still generated robust increases in 'wanting'. We also report a novel hedonic hotspot in caudal anterior cingulate cortex (pACC) never previously described, and find that ChR2 stimulation of neurons in that pACC hotspot increases hedonic 'liking' as well as incentive motivation or 'wanting' to consume sucrose. Subcortically, we show that VP neurons bidirectionally control hedonic impact, as optogenetic inhibitions of the VP hotspot decreased hedonic 'liking' reactions to sweet sucrose and replaced them with aversive 'disgust' reactions. We further selectively targeted VP^{GABA} neurons using transgenic GAD1-cre rats, and show that specific optogenetic activation of VP^{GABA} neurons within the posterior VP hotspot doubles hedonic 'liking' reactions to sucrose. By contrast, rostral VP^{GABA} activations failed to increase hedonic 'liking' reactions to sucrose, but still 'wanting' to consume sucrose, and other motivational 'wanting' effects such as robust laser self-stimulation and can even maladaptive attraction to a laser-paired electric shock rod. Patterns of distributed Fos expression in distant brain structures following optogenetic stimulation of a hedonic hotspot suggested that hedonic hotspot stimulation recruits widespread mesocorticolimbic systems to enhance 'liking'. Overall, our findings suggest that hedonic hotspots in the brain are robust hedonic entities in brain corticolimbic sites, whether stimulated pharmacologically or optogenetically, that form a functional hedonic circuit to enhance 'liking'.

Disclosures: I. Morales: None. **J. Kravchenko:** None. **K.E. Emery:** None. **M. Ayoub:** None. **K.R. Urstadt:** None. **D.C. Castro:** None. **K.C. Berridge:** None.

Poster

PSTR165. Motivation and Food

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR165.02/LL20

Topic: G.03. Motivation

Support: NIH NIDDK Grant R01DK085721

Title: Chemogenetic manipulations of CRH neurons in the central amygdala impact habituation to novel palatable food differently in male and female rats

Authors: *Z. R. IRVING, R. SHTEYN, G. D. PETROVICH;
Psychology and Neurosci., Boston Col., Boston, MA

Abstract: Environmental and physiological factors influence responses to novel stimuli. Food consumption is carefully regulated under novelty due to potential dangers from unfamiliar foods and places. Our prior work found that rats habituate to novel food differently when tested in a novel versus familiar contexts, and there were sex differences. Corticotropin-releasing hormone/factor (CRH/CRF) neurons are involved in stress regulation and appetitive motivation. Here, we examined the role of CRH neurons in the central nucleus of the amygdala (CEA) in habituation to a novel food and context using DREADDs. Adult male and female CRH-Cre rats received viral infusions in the CEA (histology pending) of either Gq-coupled DREADDs to activate CRH neurons, Gi-coupled DREADDs to inhibit CRH neurons, or a control (DIO-mCherry) virus. Rats were food deprived for 20h prior to each of the 4 habituation sessions in either in a novel or familiar context. During each session, rats were given a choice of *novel*, palatable Test Diet (TD) pellets and their *familiar* standard rat chow. DREADD ligand J60 or saline was administered prior to sessions 1 and 3. Control Gq and Gi groups received saline, while control virus groups received J60. All controls were pooled in one group for the current analysis. Consumption patterns were analyzed by grams eaten per 100 grams of body weight. During the first session, rats consumed less of the novel food, and Gq groups consumed more chow than Gi groups ($p<0.05$). In the novel context, there were no differences in consumption between viral groups, and TD was consumed more than chow ($p=0.006$). During habituation 2, when DREADDs were not activated, there were no differences between viral groups in either context ($p>0.05$). During habituation 3, when DREADDs were activated for the second time, in the familiar context, females and Gq groups consumed more of both foods ($p<0.05$). In the novel context, viral groups did not differ, though females still had greater consumption ($p=0.087$). During the final habituation, when DREADDs were not activated, viral groups did not differ in either context, however there were interactions between food type, virus group, and sex in both contexts (novel $p=0.081$, familiar $p=0.034$). Female Gq groups tested in either context consumed more TD than males ($p<0.01$). Male Gi rats tested in the familiar context consumed more TD than females ($p=0.005$) but in the novel context, Gi rats of both sexes consumed similar amounts of TD ($p=0.368$). These results provide preliminary evidence that CRH neurons in the CEA are relevant to habituation to a novel, palatable food. Excitation of CRH neurons appears to manipulate the hedonic value of food, and more so in females.

Disclosures: Z.R. Irving: None. **R. Shteyn:** None. **G.D. Petrovich:** None.

Poster

PSTR165. Motivation and Food

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR165.03/LL21

Topic: G.03. Motivation

Support: NIH NIDDK Grant RO1 DK085721

Title: Chemogenetic manipulations of CRH neurons in central amygdala impact familiar palatable food consumption differently in male and female rats

Authors: *R. SHTEYN, Z. R. IRVING, G. D. PETROVICH;
Boston Col., Boston, MA

Abstract: Food palatability can drive eating in the absence of hunger. Our group recently found that female rats are particularly sensitive to consuming palatable food when they are sated, unlike males, who eat in line with their hunger state. The central nucleus of the amygdala (CEA) is critical in processing motivational stimuli related to feeding, stress, and both natural and drug rewards, and may underlie these hedonic feeding sex differences. Of particular interest within the CEA are corticotropin-releasing hormone/factor (CRH/CRF)-expressing neurons, which regulate stress responses, but have also been implicated in appetitive events, such as feeding. Here we examined the role of CEA^{CRH} neurons in regulating palatable food (PF; Test Diet pellets) intake. Adult male and female CRH-Cre rats received bilateral injections of excitatory (Gq-coupled), inhibitory (Gi-coupled), or control (DIO-mCherry) DREADD virus into the CEA (histology pending). Following 3-week viral expression, these subjects participated in another experiment (Irving, Shteyn, and Petrovich, 2023 SfN abstract) where they were habituated to the PF. Then they underwent a Pavlovian cue-food learning protocol and food consumption tests with the PF. Thus, by the time of consumption testing, the subjects had considerable experience with the PF. Testing occurred in a familiar environment (home cage) and compared rats' preference for the PF vs. neutral-tasting standard chow. Sated rats were tested twice, once with the PF and once with chow (test order counterbalanced). 30 minutes prior to testing, DREADD selective ligand J60 (JHU37160; 0.2mg/kg) was administered to selectively activate or inhibit CEA^{CRH} neurons. Control Gq and Gi groups received saline, while control virus groups received J60. All controls were pooled in one group for the current analysis. Preliminary consumption test patterns, analyzed by grams eaten per 100g of body weight, showed that all rats ate more TD than chow ($F(1, 25)=233.161$, $p<.001$), and that females ate more than males ($F(1, 25)=13.191$, $p=.001$). The excitatory and inhibitory DREADD groups ate slightly more than the control group ($F(1, 25)=2.808$, $p=.079$), but did not differ from each other ($p=.100$). Females ate more than males in the excitatory and inhibitory but not control group ($p=.004$, $p=.002$, $p=.840$). These preliminary data replicate our prior findings that sated females are more prone to eating palatable food than males. They also suggest that CEA^{CRH} neurons play a sex-specific role during hedonic feeding, and intriguingly, that both exciting and inhibiting CEA^{CRH} neurons stimulates this hedonic PF consumption.

Disclosures: R. Shteyn: None. **Z.R. Irving:** None. **G.D. Petrovich:** None.

Poster

PSTR165. Motivation and Food

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR165.04/LL22

Topic: G.03. Motivation

Support: NIDA Grant R00045765
NJHF Grant PC98-22
Busch Biomedical Research Grant (FP00039540)
NSF Award HRD-1909824

Title: Sigma 1 receptor antagonism preferentially reduces binge-like eating and motivated responding for palatable food in female rats maintained on a high fat diet

Authors: *A. J. ARMANIOUS[1,3], J. E. FINLEY[1,3], Y. PENG[4], W. J. WELSH[2,4], M. H. JAMES[1,3];
[1]Psychiatry, [2]Pharmacol., Robert Wood Johnson Med. School, Rutgers Univ., Piscataway, NJ; [3]Brain Hlth. Institute, Rutgers Univ., Piscataway, NJ; [4]Rutgers-Cancer Inst. of New Jersey, Rutgers Univ., New Brunswick, NJ

Abstract: Introduction: Binge eating disorder (BED) is disproportionately observed in persons of higher weight. Despite this, preclinical studies examining novel pharmaceutical treatments for BED are typically carried out in lean rats. Sigma-1 receptor (S1R) antagonists are effective at reducing motivated food seeking in lean rats, however their efficacy has not been tested in a rat model of obesity. Here, we sought to test if a novel, highly potent sigma-1 receptor (S1R) antagonist (PW507), differentially alters binge-like eating in rats maintained on a high fat diet (HFD) vs. lean rats. **Methods:** Female Long Evans rats were maintained on either a regular chow diet (15% fat, ad libitum; n=8) or a HFD (45% fat, ad libitum; n=16) for 8w before being given restricted, intermittent access (30 min, twice/week, 4w) to sweetened fat (vegetable shortening/10% sucrose) to promote binge-like eating. Rats were then trained to lever press for sucrose pellets on a low (fixed ratio [FR] 1) and high effort (FR5) schedules of reinforcement, followed by testing on a progressive ratio schedule. Rats were treated with PW507 (0, 5, 10, 15, 20mg/kg; i.p.) prior to binge and operant test sessions (within-subjects, counterbalanced). We also tested the effect of PW507 on general locomotor activity. **Results:** In HFD rats, PW507 dose-dependently decreased binge-like eating, FR5 sucrose responding and PR breakpoints; no such effects were observed in lean rats. At the lowest effective dose (10mg/kg), PW507 had no effect on low effort (FR1) responding for sucrose or general locomotor activity in both HFD and lean rats. **Conclusions:** Blocking S1R reduced binge-like eating and motivated food responding in HFD but not chow rats, indicating that these behaviors become S1R-dependent only under conditions of diet-induced obesity. These data underscore the importance of testing potential therapeutics for BED and associated conditions using animal models that recapitulate the weight status of clinical populations.

Disclosures: A.J. Armanious: None. **J.E. Finley:** None. **Y. Peng:** None. **W.J. Welsh:** None. **M.H. James:** None.

Poster

PSTR165. Motivation and Food

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR165.05/LL23

Topic: G.03. Motivation

Support: NIH T34GM145384

Title: Effects of adolescent sucrose exposure on operant novelty seeking in male and female rats

Authors: *N. ALAMMARI[1], B. KRIEG[2], I. GARCIA[1], S. SOTO[1], A. M. GANCARZ[1];
[2]California State University, Bakersfield, [1]California State University, Bakersfield, Bakersfield, CA

Abstract: In humans, adolescence is a transitional period between childhood and adulthood and is accompanied by significant behavioral and neural development. Environmental challenges during this crucial time of neural plasticity could potentially result in dire neurodevelopment and behavior consequences in adulthood. It is known that adolescent exposure to drugs of abuse increases future susceptibility to drug addiction in adulthood, yet little is known about the effects of exposure to other natural stimuli on drug abuse in adulthood. Given that natural and drug reinforcers activate the same neural circuitry, this pathway may also be responsible for such increases in reactivity to natural reinforcers (e.g., sucrose) during the adolescent period.Sensation Seeking (SS) is a personality trait that has been linked with drug addiction and is generally described as the preference for novel sensations and experiences. In humans, high SS scores are associated with a greater likelihood of drug abuse. In rodents, operant novelty seeking (ONS), an animal model of Sensation Seeking, has been shown to be predictive of acquisition of drug self-administration and is operationally defined as operant responding maintained by varied, novel auditory/visual stimuli. The primary goal of this research is to evaluate the hypothesis that exposure to sucrose in adolescence alters sensitivity to the reinforcing effects of novel visual/auditory stimuli in adulthood.We seek to understand how exposure to sucrose could alter behavior in adulthood. To test this, male and female adolescent rats were exposed to either 10% sucrose or water in fifteen 2-hour sessions. In adulthood, rats were first habituated to dark experimental chambers and then tested on the ONS task where active responses were reinforced according to a VI 1-min reinforcement schedule. Reinforced responses resulted in a complex visual/auditory stimulus, which consisted of the illumination of stimulus lights of various colors and the presentation of an auditory stimulus. Preliminary results indicate females preexposed to sucrose emitted significantly more responses during habituation, when no sensory stimuli were available, compared to water controls. However, contrary to our hypothesis, this effect dissipated upon the introduction of the response contingent sensory reinforcers. Performance remained sexually dimorphic in the ONS phase, with females emitting significantly more active responses and earning more sensory rewards than their male counterparts. These data indicate adolescent preexposure to sucrose may produce sex-specific differences in generalized exploratory behavior in adulthood.

Disclosures: **N. Alammari:** None. **B. Krieg:** None. **I. Garcia:** None. **S. Soto:** None. **A.M. Gancarz:** None.

Poster

PSTR165. Motivation and Food

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR165.06/LL24

Topic: G.03. Motivation

Support: NIH Grant T34GM145384

Title: Adolescent Sucrose Exposure Alters Operant Responding for Sucrose in Adulthood

Authors: B. KRIEG[1], K. HIGDON[1], K. BANDUCCI[2], B. REED[1], I. GARCIA[1], S. SOTO[1], N. ALAMMARI[1], *A. GANCARZ-KAUSCH[3];
[1]Dept. of Psychology, [2]Dept. of Biol., [3]California State University, Bakersfield, Bakersfield, CA

Abstract: An issue that affects many individuals in Western societies is excessive sugar consumption, which has been shown to have both short- and long-term adverse effects on health. Adolescents are at particular risk due to the increased plasticity of the brain during that developmental period. Therefore, it important to study the effects of adolescent sucrose exposure on consumption of sucrose throughout the lifespan. We hypothesized that chronic sucrose exposure during adolescence would alter sucrose reinforcement in adulthood. To test this, female and male Sprague Dawley rats were given short access (2 h/day) to either 10% sucrose or water in their homecages each day during the adolescent period (PND28 - 42). Following adolescence, rats were left undisturbed with access to water until adulthood. Once rats reached adulthood, they were tested for 2-hour operant self-administration paradigm in which they earned 10% sucrose solution (100 µl/presentation) according to an FR1 schedule of reinforcement for 10 days. Rats were then tested for motivation to work for sucrose via a single progressive ratio test. Rats had 5 hours to emit increasing response requirements for successive presentations of sucrose solution. Failure to emit the required responses in 1 hr was termed a 'breakpoint' and resulted in termination of the session. Preliminary results indicate significant main effects of preexposure, whereby rats pre-exposed to sucrose in adolescence emitted fewer active, but not inactive, responses, leading to fewer sucrose reinforcers earned compared to water controls. However, no significant difference in performance on a progressive ratio schedule was observed. Furthermore, a significant main effect of sex was observed, in which females emitted more active, but not inactive, responses and earned more sucrose solution across days of testing on an FR1 schedule and achieved significantly higher breakpoints compared to male counterparts. Results indicated that adolescent exposure to sucrose decreased propensity for operant responding for sugar in adulthood. One potential interpretation is that sucrose in adolescence produces anhedonia and decreases the hedonic value of the sucrose in adulthood. Future research is needed to fully explore the ontogeny of sucrose vulnerability.

Disclosures: **B. Krieg:** None. **K. Higdon:** None. **K. Banducci:** None. **B. Reed:** None. **I. Garcia:** None. **S. Soto:** None. **N. Alammari:** None. **A. Gancarz-Kausch:** None.

Poster

PSTR165. Motivation and Food

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR165.07/LL25

Topic: G.03. Motivation

Title: Adolescent Oxycodone Exposure Impacts Motivation for a Sucrose Reward

Authors: *C. A. SMITHSON, J. T. COOK, S. A. ROBINSON;
Williams Col. Neurosci. Program, Williamstown, MA

Abstract: Adolescent opioid exposure is experiencing a growing prevalence in the United States, as between just 2019 and 2021, the number of adolescent overdose deaths quadrupled to 2,155 deaths. Adolescent opioid exposure has been connected to an increased risk of developing substance-use disorders, which is attributed to dysregulation in the mesolimbic dopamine system. Other research studies have focused around the effects of opioids on the system in adult mice, however our lab was interested in understanding the effect going into adulthood after an adolescent exposure. Beginning at 5 weeks old, male C57/BL6 mice were exposed to bi-daily escalating doses of oxycodone (9 mg/kg - 33 mg/kg) or saline for a period of 5 days (n = 10/group), followed by one week of abstinence. During the abstinence period, an open field test was performed at 8 hrs, 24 hrs, 48 hrs, 72 hrs, and 1 week following the last injection of oxycodone or saline to evaluate if any anxiety or lethargy phenotypes were present. The total distance traveled and the time spent in the inner and outer zones was measured using Anymaze software. A repeated measures ANOVA demonstrated a significant time x drug interaction ($p=0.0008$) in the total distance traveled and a multiple comparisons analysis revealed that the oxycodone mice were more lethargic than saline controls at 48 hrs ($p=0.0482$) and 1 week ($p=0.0095$) after the last injection of oxycodone. There also was a significant time x drug interaction ($p =0.0343$) in the amount of time spent in the inner zone, but the analysis yielded no statistically significant multiple comparisons. After 1 week of abstinence, all mice underwent an operant food reward task in which they had to learn to nose-poke at an active port to receive a sucrose pellet reward using a progressive ratio schedule. Oxycodone-exposed mice performed more active nosepokes during the progressive ratio timepoint (in which there are exponentially increasing requirements for a single pellet) compared to saline treated mice, demonstrating an increased motivation for a sucrose reward ($p<0.001$). Future experimentation will evaluate the role of neural plasticity in modulating increased motivation for a sucrose reward in the mesolimbic pathway. The mRNA expression of BDNF, CREB, and GABA receptor alpha-subunit 1 will be measured in the ventral tegmental area, nucleus accumbens, and the prefrontal cortex using quantitative real-time polymerase chain reaction.

Disclosures: **C.A. Smithson:** None. **J.T. Cook:** None. **S.A. Robinson:** None.

Poster

PSTR165. Motivation and Food

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR165.08/LL26

Topic: G.03. Motivation

Support: NRSA F31DA057112-0

Title: Contributions of estradiol and progesterone to female risk aversion

Authors: *L. M. TRUCKENBROD[1], M. GARNER[2], N. R. CARLOS[3], A. C. GORE[4], C. A. ORSINI[5];
[1]Neurosci., Univ. of Texas, Austin, TX; [2]Neurol., Univ. of Texas at Austin, Austin TX, TX; [3]Psychology, [4]Univ. of Texas at Austin, Univ. of Texas at Austin, Austin, TX; [5]UT Dell Med. Sch., The Univ. of Texas at Austin, AUSTIN, TX

Abstract: Decision making is a complex cognitive process in which an individual must weigh options that differ in their expected rewards and their associated costs. Previous literature has established sex differences in decision making, particularly when decisions involve an explicit risk of punishment, with females displaying greater risk aversion than males. Recent evidence suggests that the ovarian hormone estradiol is a critical mediator of phenotypical female risk aversion. The role of progesterone in this form of decision making, however, is unknown. To address this gap in knowledge, female Long-Evans rats (n=19) were trained in a model of risk-based decision making (the Risky Decision-making Task, RDT) in which rats choose between a small, safe food reward and a large food reward that is accompanied by a variable probability of footshock punishment. After achieving stable behavioral performance, rats were ovariectomized (OVX) and re-tested in the RDT. Rats then received subcutaneous administration of estradiol benzoate (EB, 0.05mg/kg), EB and progesterone (EB+P; EB, 0.05mg/kg; P, 0.5mg/kg) or vehicle (sesame oil) daily for 7 days. Injections occurred following testing in the RDT each day using a randomized within-subjects design, such that each rat received 7 days of each treatment with a minimum of 8 days between treatments. During each treatment and the successive washout period, the rats' estrous cycles were assessed to confirm that the hormonal state of the rat was consistent with their treatment group. Consistent with previous work, OVX increased risk taking relative to rats' pre-surgical performance. Administration of either EB or EB+P attenuated this effect, causing a decrease in risk taking. To determine whether the decrease in risk taking in the EB+P condition was due to EB alone, rats underwent another regimen of hormone treatment in which P (or vehicle) was administered alone (identical duration and dosing as above). In contrast to the effects of EB alone, there were no effects of P on risk taking. These data expand our understanding of hormonal regulation of risk taking and indicate that estradiol is the critical ovarian hormone responsible for female risk aversion.

Disclosures: L.M. Truckenbrod: None. **M. Garner:** None. **N.R. Carlos:** None. **A.C. Gore:** None. **C.A. Orsini:** None.

Poster

PSTR165. Motivation and Food

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR165.09/LL27

Topic: G.03. Motivation

Title: Chronic caffeine administration reduces fluoxetine-induced sexual dysfunction in female Long-Evans rats

Authors: *M. A. KELLY, F. A. GUARRACI, R. SANDERCOCK, K. MYERS; Southwestern Univ., Georgetown, TX

Abstract: The current study examined the effects of chronic caffeine administration on sexual dysfunction caused by an acute dose of fluoxetine in female Long-Evans rats. Previous research has highlighted the increasing prevalence of chronic caffeine consumption in humans as well as the high rate of sexual dysfunction in patients taking SSRIs, such as fluoxetine (Fisonen et al., 2004; Baldwin, 2015). Studies have found that chronic caffeine administration is linked to increased sexual motivation in rats (Guarraci & Benson, 2005), but little is known about how the interaction between fluoxetine and chronic caffeine consumption affects sexual behavior. We hypothesized that chronic caffeine administration followed by a combined dose of caffeine and fluoxetine could protect against fluoxetine-induced sexual dysfunction. Subjects consisted of 14 ovariectomized female Long-Evans rats that were randomly assigned to receive either caffeine (15mg/kg) or saline (15mg/kg) through an intraperitoneal (i.p.) injection for 8 days. On the 8th day of the experiment, in addition to receiving caffeine or saline, all rats received a dose of fluoxetine (10 mg/kg; i.p.) 30 minutes before assessing sexual motivation using the partner-preference paradigm. In order to induce behavioral estrus and sexual receptivity during the partner-preference tests, all female rats were primed with 10.0 μg of estradiol benzoate 48 hours and 1.0 mg of progesterone 4 hours prior to testing. During partner-preference testing, subjects were given the opportunity to approach either a sexually vigorous opposite-sex stimulus or a same-sex stimulus to assess sexual motivation. The results of an independent samples *t*-test supported our hypothesis and found that subjects treated with a combination of caffeine and fluoxetine spent significantly more time with the male stimulus and visited the male stimulus more frequently compared to subjects treated with a combination of saline and fluoxetine. Therefore, we concluded that chronic caffeine administration combined with an acute dose of fluoxetine significantly increased sexual motivation compared to controls and protected against fluoxetine-induced sexual dysfunction. The results of this study provide valuable insight that could explain why some people experience fluoxetine-induced sexual dysfunction whereas others are unaffected by this side effect.

Disclosures: M.A. Kelly: None. **F.A. Guarraci:** None. **R. Sandercock:** None. **K. Myers:** None.

Poster

PSTR165. Motivation and Food

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR165.10/Web Only

Topic: G.03. Motivation

Support: CONACYT Grant 720657

Title: Effects of the exposure to a hypercaloric diet on incentive motivation in rats

Authors: *W. ZEPEDA-RUIZ[1], D. N. VELÁZQUEZ-MARTÍNEZ[2];
[1]Univ. Nacional Autonoma De Mexico, Mexico, Mexico; [2]Univ. Nacional Autónoma de México, Inst. Nacional de Perinatología, Facultad de Psicología, Mexico

Abstract: Hypercaloric diets have been related to motivational changes, some authors have reported a decreased motivation to obtain palatable food after exposure to high-fat or high-sucrose diets; however, others have reported an increased motivation. Since the effects of the hypercaloric diet appear to depend on the time of exposure to the diet, we wonder if a prolonged exposure to the hypercaloric diet produces long-lasting effects on motivation. Therefore, the objective of the present work was to evaluate the performance of the subjects on the progressive ratio schedule during twenty weeks of access to a high-fat, high-sucrose choice (HFHSc) diet. Twenty-four male Wistar rats (250-300 g) were trained in a progressive ratio schedule. After achieving behavioral stability, subjects were divided in two groups (control and experimental) according to their breaking point in a counterbalanced way. During the exposure to the HFHSc diet, subjects of both groups had *ad libitum* access to a standard diet and water, in addition, subjects of the experimental group had free access to a 10% sucrose solution and edible fat (INCA®). Throughout 20 weeks, subjects were maintained in their respective diets and were evaluated in the progressive ratio schedule. We found that, after two weeks of exposure to the hypercaloric diet, subjects of the experimental group achieved lower breaking points in comparison to the control group and to the breaking point achieved during the training phase. From the third week of exposure to the hypercaloric diet differences disappeared and we did not find significant differences between groups when the breaking point was compared. In addition, we measured glucose levels in fasting conditions and the weight of perigonadal, peritoneal and retroperitoneal adipose tissue. After 20 weeks of access to the HFHSc diet, subjects of the experimental group had elevated glucose levels, and the weight of the adipose tissue was larger in comparison with the control group. Our results suggest that it is not necessary to employ long periods of diet exposure to evaluate its effects on motivation, because the decreased in motivation in the experimental group was observed during the first two weeks of diet exposure. Worth of notice, the HFHSc diet replicates some characteristics of obesity in humans, such as elevated glucose levels in fasting conditions and an increase in the adipose tissue weight.

Disclosures: W. Zepeda-Ruiz: None. **D.N. Velázquez-Martínez:** None.

Poster

PSTR165. Motivation and Food

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR165.11/LL28

Topic: G.03. Motivation

Support: R15 DA016285-05

Title: Effects of unrestricted sweet access on sucrose cue reactivity in adult female and male rats

Authors: *J. GRIMM, C. BACON, J. BARSON, T. FROST-BELANSKY, F. HERNANDEZ-ROMAN, L. JACKSON, K. SANKO, J. WEBER;
Western Washington Univ., Bellingham, WA

Abstract: We previously found that unrestricted access to sucrose solution (“satiation”) reduces sucrose cue-reactivity in male rats, but only in early abstinence from sucrose self-administration. The purpose of the present study was to examine whether this relationship generalizes to the non-nutritive sweetener, saccharin, and to female rats. Adult rats lever pressed for 0.2 mL 10% sucrose in 10, 2 h daily sessions. A tone+light cue was presented along with every reinforced response. Rats were then randomly assigned to be tested in either early (2 days) or late (32 days) abstinence from their 10th day of sucrose self-administration. For the 46 h prior to testing, rats were satiated in home cages with either drinking water, 10% sucrose, or .3% saccharin. As described in previous works, female rats responded at a higher rate for sucrose and for the sucrose-paired cue. In addition, sucrose cue-reactivity was greater in late vs. early abstinence ("incubation of craving"). Both male and female rats consumed more sweet solution (sucrose or saccharin) than water during the 46-h satiation manipulation. This sweet satiation reduced subsequent sucrose cue-reactivity in early but not late abstinence. These findings replicate our previous findings where prolonged access to sucrose had no effect on the incubation of sucrose craving. Furthermore, this effect generalizes to the non-nutritive sweetener saccharin and to female rats. Essentially, the ability of a primary reinforcer or similar (sucrose, saccharin) to reduce conditioned responding is diminished after a period of abstinence from chronic intake. That is, in prolonged abstinence a sucrose-paired cue will continue to guide behavior even after the individual has consumed a large amount of sweet fluid. One implication is that unrestricted access to sweet will not satiate conditioned craving for sucrose following several weeks of abstinence from sucrose.

Disclosures: J. Grimm: None. **C. Bacon:** None. **J. Barson:** None. **T. Frost-Belansky:** None. **F. Hernandez-Roman:** None. **L. Jackson:** None. **K. Sanko:** None. **J. Weber:** None.

Poster

PSTR165. Motivation and Food

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR165.12/MM1

Topic: G.03. Motivation

Title: Hypothalamic neuronal activation in non-human primates drives naturalistic goal-directed eating behaviour

Authors: *L. HA[1], H.-G. YEO[2], Y. KIM[2], I. BAEK[1], E. BAEG[3], J. CHOI[4], J.-H. CHO[5], Y. LEE[2], H. CHOI[1];
[1]Seoul Natl. Univ., Seoul, Korea, Republic of; [2]Korea Res. Inst. of Biosci. and Biotech. (KRIBB), Cheongju, Korea, Republic of; [3]CNIR, Inst. For Basic Sci. (IBS), CNIR, Inst. For Basic Sci. (IBS), Suwon, Korea, Republic of; [4]Korea Inst. of Radiological and Med. Sci., Korea Inst. of Radiological and Med. Sci., Seoul, Korea, Republic of; [5]Bio-Chemical Analysis Team, Korea Basic Sci. Inst., Cheongju, Korea, Republic of

Abstract: Eating addiction is the primary cause of modern obesity. Although the causal role of the lateral hypothalamic area (LHA) in eating has been demonstrated in rodents, there is no evidence in non-human primates regarding naturalistic eating behaviours. Therefore, we investigated the role of LHA GABAergic (LHA^{GABA}) neurons in eating in three macaques using chemogenetics, a novel drug-controlled neuromodulation gene therapy. LHA^{GABA} neuron activation significantly increased naturalistic goal-directed behaviours and food motivation specific to palatable food. Positron emission tomography and magnetic resonance spectroscopy validated the chemogenetic activation. Resting-state functional magnetic resonance imaging showed that functional connectivity (FC) between the LHA and frontal areas increased, while the FC between the frontal cortices decreased after LHA^{GABA} neuron activation. Thus, our study elucidates the role of LHA^{GABA} neurons in obesity. Moreover, the current preclinical proof-of-concept evidence suggests the use of novel chemogenetic gene therapy for the treatment of obesity and neuropsychiatric disorders.
Keyword: Non-human primate, Eating behavior, Obesity, Chemogenetic, Goal-directed behavior

Disclosures: L. Ha: None. **H. Yeo:** None. **Y. Kim:** None. **I. Baek:** None. **E. Baeg:** None. **J. Choi:** None. **J. Cho:** None. **Y. Lee:** None. **H. Choi:** None.

Poster

PSTR165. Motivation and Food

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR165.13/MM2

Topic: G.03. Motivation

Support: STI 2030—Major Projects 2021ZD0200500
Shanghai Municipal Natural Science Foundation (21ZR1419700)

Title: Glutamatergic input from lateral parabrachial nucleus to ventral tegmental area inhibits dopamine neurons via BK channel in activity based anorexia

Authors: B. PENG, Y. CHEN, X. GAO, Y. XIN, Y. ZHANG, D. YANG, C. BAO, ***S. LIU**; East China Normal Univ., Shanghai, China

Abstract: Anorexia nervosa (AN) is a psychiatric disease with high mortality. The compulsivity of the disorder leads to an emerging dopamine-centered hypothesis, with a growing body of literature showing the involvement of the dopaminergic system. However, the key underlying mechanism for AN is still elusive. In the present study, we investigated the role of the lateral parabrachial nucleus (LPBN) to the ventral tegmental area (VTA) circuit, in a well-established animal model of AN (activity-based anorexia, ABA). Using in vivo and in vitro electrophysiology, optogenetics, chemogenetics, and fiber photometry, we found that manipulating the LPBN-VTA circuit toggles the food intake and reinforcement behaviors. Besides, the excitability of VTA-projecting LPBN neurons was increased in ABA mice, and inhibition of this circuit significantly increased ABA mice's food intake and survival rate. The LPBN glutamatergic neurons targeted the medial-VTA DA neurons via indirect inhibitory GABA interneurons, which resulted in the hypoactivity of the VTA DA neurons in ABA mice. This hypoactivity concurred with the decreased function of the large conductance, calcium- and voltage-activated potassium (BK) channel on the DA neurons which its agonist was capable to reverse. Notably, microinjection of BK agonist into the VTA rescued DA neuron firing and survival rate in ABA mice. These results have elucidated the critical role of the LPBN-VTA circuit and the potential channel pathology in the disease, which may serve as a key to developing drug treatments and intervention strategies for AN.

Disclosures: B. Peng: None. **Y. Chen:** None. **X. Gao:** None. **Y. Xin:** None. **Y. Zhang:** None. **D. Yang:** None. **C. Bao:** None. **S. Liu:** None.

Poster

PSTR165. Motivation and Food

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR165.14/MM3

Topic: G.01. Fear and Aversive Learning and Memory

Support: Helen Hay Whitney Foundation
Simons Collaboration on the Global Brain
Brain Research Foundation
NIH Grant U19-NS123716
NIH Grant U19-NS104648

Title: Visceral feedback signals reactivate neural flavor representations during postingestive learning

Authors: *C. A. ZIMMERMAN, A. PAN-VAZQUEZ, B. WU, E. F. KEPPLER, R. N. FETCHO, S. S. BOLKAN, B. MCMANNON, E. M. GUTHMAN, J. LEE, A. HOAG, L. A. LYNCH, S. N. JANARTHANAN, A. G. BONDY, S. S. H. WANG, I. B. WITTEN; Princeton Univ., Princeton, NJ

Abstract: Animals learn the value of foods based on their postingestive effects, and thereby develop preferences for foods that are nutrient-rich and aversions towards those that produce illness. However, it remains unclear how the brain is able to associate flavors experienced during a meal with visceral feedback signals that arise after a delay of minutes or hours. To gain insight into this temporal credit assignment problem, we leveraged the fact that mice learn to associate novel — but not familiar — foods with gastric malaise signals to investigate what distinguishes the neural representations of flavors that promote learning versus those that do not. Surveying brainwide expression of the immediate early gene Fos during drinking and postingestive malaise revealed that a cluster of amygdala regions, especially the central amygdala (CEA), is preferentially activated when mice consume novel flavors that permit learning. Moreover, this novelty-specific activation is amplified by the arrival of malaise signals from the gut. These Fos observations suggest that malaise signals may specifically reactivate flavor-coding neurons in the CEA in order to promote plasticity and postingestive learning. To test this hypothesis, we used chronic high-density Neuropixels recordings to track the activity of individual CEA units across drinking and malaise. This revealed that malaise signals preferentially reactivate novel flavor-coding CEA units versus other nearby cells, and that population dynamics during bouts of malaise closely mirror those during novel flavor drinking. Neurons that were strongly reactivated by malaise showed strengthened flavor responses during a memory retrieval test the following day, which suggests that postingestive reactivations serve to trigger relevant plasticity of CEA representations. Together, our findings demonstrate that reactivation of neural flavor representations can bridge the gap between drinking and delayed visceral feedback, suggesting a novel mechanism to support postingestive learning.

Disclosures: C.A. Zimmerman: None. **A. Pan-Vazquez:** None. **B. Wu:** None. **E.F. Keppler:** None. **R.N. Fetcho:** None. **S.S. Bolkan:** None. **B. McMannon:** None. **E.M. Guthman:** None. **J. Lee:** None. **A. Hoag:** None. **L.A. Lynch:** None. **S.N. Janarthanan:** None. **A.G. Bondy:** None. **S.S.H. Wang:** None. **I.B. Witten:** None.

Poster

PSTR165. Motivation and Food

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR165.15/Web Only

Topic: G.03. Motivation

Support: UNAM-PAPIIT IN232120

Title: Systemic activation of CB2 receptors decreases motivation for palatable food in subjects with diet-induced obesity

Authors: V. FRANCO-QUIROZ[1], I. CONDE-ROJAS[2], F. CORTÉS-SALAZAR[3], V. LÓPEZ-ALONSO[3], J. MANCILLA-DÍAZ[3], ***R. ESCARTIN-PEREZ**[3];
[1]Maestría en Ciencias (Neurobiología), [2]Posdoc DGAPA, [3]División de Investigación y Posgrado, UNAM FES Iztacala, Tlalnepantla, Mexico

Abstract: The study of the neurobiology of obesity has focused on the brain mechanisms that control eating patterns to understand the neural processes that contribute to abnormal weight gain. Obesity causes multiple changes in the functioning of the central nervous system, including increased circulating endocannabinoid concentrations, alterations in CB1 receptor expression, and behavioral changes that promote even more weight gain. Brain regions where the activity of reward and emotional regulation circuits converge have in common that cannabinoid transmission has significant neuromodulatory activity. CB1 receptors (CB1r) are known to stimulate the consumption of palatable food; however, the contributions of CB2 receptors (CB2r) in eating behavior are not yet fully clarified. There are only a few reports that CB2r activation produces contrasting effects to those produced by CB1r activation. Accordingly, we assessed the effects of systemic CB2r activation on motivation for palatable food using an operant task in diet-induced obese rats. We fed male Wistar rats (140-160 g) with a high-fat diet (HFD, 60% of energy from fat, n= 10) or the control diet (CD, similar flavor, 10% of energy from fat, n=10) for 64 days. During the exposure to the HFD, the animals were trained with a sequence of operant conditioning programs, starting with a fixed ratio 1 (FR1), then with a fixed ratio 5 (FR5), and finally with a progressive ratio program (RP, response ratio= [5e(0.2 × trial number)] - 5) to measure motivation for chocolate-flavored sucrose pellets. We measured biochemical markers in plasma (cholesterol, triglycerides, and glucose), weight gain (g), and the fat mass/body weight ratio to validate the obesity model. We assessed motivation for palatable food before and after the pharmacological activation of the CB2r by measuring breakpoints under a PR schedule of reinforcement. We found that animals exposed to HFD gained significantly more body weight and accumulated more fat mass. Also, the levels of cholesterol (LDL) and triglycerides in the plasma of DIO rats were higher than controls after two months of HFD consumption. According to the behavioral tests, breakpoints of rats exposed to HFD were significantly lower than controls before the pharmacological tests, and the activation of CB2r with GW405833 (3 mg/kg) significantly decreased breakpoints. This effect was prevented by the CB2r antagonist (AM630 10 mg/kg). Our results are consistent with the hypothesis that CB2r play an opposite role to CB1r, as their activation decreases the motivation for palatable food, especially under conditions of diet-induced obesity.

Disclosures: V. Franco-Quiroz: None. **I. Conde-Rojas:** None. **F. Cortés-Salazar:** None. **V. López-Alonso:** None. **J. Mancilla-Díaz:** None. **R. Escartin-Perez:** None.

Poster

PSTR165. Motivation and Food

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR165.16/MM4

Topic: G.03. Motivation

Support: R01 DK131441

Title: Serotonin regulates paraventricular thalamus neurons to modulate food intake

Authors: *Q. YE, J. NUNEZ, X. ZHANG;
Florida State Univ., Tallahassee, FL

Abstract: Central serotonin (5-HT) signaling is known to play critical roles in the regulation of food intake and eating disorders. Paraventricular thalamus (PVT) is a brain region that regulates both food motivation and consumption through connections with brainstem, hypothalamic, and cortical areas. We recently reported that 5-HT directly and indirectly activated PVT neurons via 5-HT 7 and 1A with slice patch-clamp recordings, and photostimulation of the projections from raphe nucleus to PVT decreased motivation for food in mice. However, it remains unknown how different 5-HT receptor subtypes in this area modulate feeding behavior. Therefore, We first used RNA scope in situ hybridization to detect several 5-HT receptor subtypes and found a high-level expression of 5-HT 2C and 7 receptors but a low-level expression of 5-HT 1A receptors in PVT. Then we used a progressive ratio (PR) schedule of reinforcement combined with intracranial drug infusions to examine the role of several 5-HT receptor subtypes in PVT in regulating the motivation for food intake. We found that infusion of 5-HT, 5-CT, a 5-HT 1A, 2C, and 7 receptor agonist, and MK-212, a 5-HT 2C receptor subtype agonist, all decreased active lever presses in a PR test in fed mice. However, infusion of 8 OH-DAPT, a 5-HT 1A receptor agonist did not significantly change the breakpoints. Based on our preliminary data, we concluded that raphe nucleus sends serotonergic projections to PVT to regulate food motivation via a combination of 5-HT1A, 5-HT7, and potentially 5-HT1A receptors. In the future, we will infuse 5-HT7 receptor agonist to study feeding behavior in mice. We will also study whether high-fat-high-sucrose diet changes the modulation of 5-HT on PVT neurons and affects food intake.

Disclosures: Q. Ye: None. **J. Nunez:** None. **X. Zhang:** None.

Poster

PSTR165. Motivation and Food

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR165.17/MM5

Topic: G.03. Motivation

Support: Funds from UIUC
NARSAD Young Investigator Award
Foundation of Prader Willi Research
NIH R00 Pathway to Independence award

Title: Paraventricular thalamic MC3R circuits link energy homeostasis with anxiety-related behavior

Authors: *D. CHO, K. O'BERRY, I. POSSA-PARANHOS, J. BUTTS, N. PALANIKUMAR, P. SWEENEY;
Univ. of Illinois Urbana-Champaign, Urbana, IL

Abstract: The hypothalamic melanocortin system plays a crucial role in sensing the stored energy and communicating this information to the neural circuitry controlling motivation and emotion. This system includes “first order” agouti-related protein (AgRP) neurons and pro-opiomelanocortin (POMC) neurons located in the hypothalamic arcuate nucleus and downstream neurons containing melanocortin 3 or melanocortin 4 receptor (MC3R or MC4R). Although the function of downstream MC4R neurons has been extensively described, less is known about the identity and function of MC3R containing neurons. In this study, we utilized neuroanatomical and circuit manipulation approaches in mice and found that paraventricular thalamic (PVT) MC3R neurons are innervated by the hypothalamic AgRP and POMC neurons and are activated by various anorexigenic and aversive stimuli. Chemogenetic activation of PVT MC3R neurons increased anxiety-related behavior and reduced feeding in hungry mice, while inhibition of PVT MC3R neurons reduced anxiety-related behavior and specifically increased feeding in stressful environments. Additionally, anterograde and retrograde tracing approaches demonstrate anatomical connections between PVT MC3R cells and brain regions controlling motivation and emotion. Overall, this study outlines an important role for PVT MC3R neurons in linking the energy homeostasis with neural circuitry regulating anxiety-related behavior, representing a potential target for disorders at the intersection of metabolism and emotion, such as anorexia nervosa.

Disclosures: D. Cho: None. **K. O'Berry:** None. **I. Possa-Paranhos:** None. **J. Butts:** None. **N. Palanikumar:** None. **P. Sweeney:** None.

Poster

PSTR166. Emotion and Social Behavior in Nonhuman Mammals

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR166.01/MM6

Topic: G.04. Emotion

Title: Observational trauma promotes coping behaviors and serotonin-driven plasticity of burst activity in habenula

Authors: *S. MONDOLONI[1], P. MOLINA MOLINA[2], A. LALIVE[2], M. CONGIU[2], S. LECCA[2], Y. LI[3], M. MAMELI[2];
[1]Dept. of Neurosci., [2]The Univ. of Lausanne, Dept of Fundamental Neurosci., Univ. of Lausanne, Lausanne, Switzerland; [3]Peking Univ., Peking Univ., Beijing, China

Abstract: Life history shapes each individuals’ behavioral trait. Notably, low levels of stress heighten performance and protect from developing maladaptive behaviors after traumas. It remains however unclear whether mild negative events influence animal’s state, behavior or

neural dynamics. The lateral habenula (LHb) is a subcortical structure contributing to valence encoding and its neuronal activity adapts accordingly to the animal's affective state and after prolonged stress experience. Importantly, upregulation of burst activity in LHb neurons, defined as a transitory high-frequency of spikes, is associated to despair-like behavior after prolonged stress. While the consequence of extended stress experience onto LHb dynamics, neurotransmission and subsequent behaviors are largely explored, it remains unknown whether mild experience will have any repercussion on LHb dynamics and function. To test this, we modeled observational trauma in male mice (6-10 weeks old), where individuals witness a peer submitted to a traumatic event (foot-shocks) while control animals observed a peer not submitted to foot-shocks. We found that such experience does not modify mice basal behaviors including sociability, locomotion or reward seeking. However, observational trauma protected mice to develop stress-driven despair-like phenotype. We then used electrophysiology in brain slices and demonstrated that observational trauma reduced solely LHb burst activity. Using fiber photometry to quantify serotonin (5HT) release through the 5HT-GRAB sensor dynamics, we observed that 5HT fluorescence increased after indirect trauma. Interestingly, serotonin bath application in acute slices decreased burst activity in the LHb, a plasticity occluded after the observational trauma paradigm. In conclusion, our work provides evidence that observing a peer submitted to a trauma promotes coping behaviors in conditions of heightened stress in parallel with a downregulation of LHb burst activity through serotonin release. This supports that LHb neuronal activity adapts through emotional contagion, which may in turn have repercussions in developing despair-like behavior.

Disclosures: **S. Mondoloni:** None. **P. Molina Molina:** None. **A. Lalive:** None. **M. Congiu:** None. **S. Lecca:** None. **Y. Li:** None. **M. Mameli:** None.

Poster

PSTR166. Emotion and Social Behavior in Nonhuman Mammals

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR166.02/MM7

Topic: G.04. Emotion

Support: the National Natural Science Foundation of China (32071040 to BL, 82071241 and 81871048 to LH)
Guangdong Basic and Applied Basic Research Foundation (2023B1515040019 to BL)
Guangdong Project (2017GC010590 to BL)

Title: Sexually dimorphic cortical circuits mediate sex-specific empathic behaviors

Authors: *S. FANG, Z. LUO, Z. WEI, H. ZHANG, Y. QIN, L. HUANG, B. LI;
Dr. Sun Yat-Sen Univ., Guangzhou, China

Abstract: Empathy, the capacity to understand others' sensory or emotional state, plays a critical role in social communication. It is widely acknowledged that males and females may display different empathic responses toward the same social situation; however, the neural mechanisms underlying sex differences in empathy are largely unknown. Here we report that sexually dimorphic activation of neuronal populations and circuits in the piriform cortex (PiC) is essential for sex differences in empathy. Specifically, when witnessing a cagemate experiencing pain, female mice activate the PiC to the prelimbic cortex (PrL) pathway to mediate social preference for the cagemate in pain. In contrast, male mice activate the PiC to the medial amygdala (MeA) pathway to display excessive self-grooming. Neural circuit tracing and transcriptomic analysis revealed that the two pathways originate from two non-overlapping populations of PiC neurons with distinct transcription factor- and sex hormones-regulated gene expression signatures. These results indicate that sex differences in empathy are controlled by sexually dimorphic mechanisms at the molecular, cellular, and circuit levels. They also provide a framework for understanding the neural mechanisms underlying the deficits of empathy in many neuropsychiatric disorders.

Disclosures: S. Fang: None. **Z. Luo:** None. **Z. Wei:** None. **H. Zhang:** None. **Y. Qin:** None. **L. Huang:** None. **B. Li:** None.

Poster

PSTR166. Emotion and Social Behavior in Nonhuman Mammals

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR166.03/MM8

Topic: G.04. Emotion

Support: NIH Grant AG051510
VA Gant BX005642
CWRU funds

Title: An insular-amygdala circuit gates familiarity-dependent observational fear

Authors: *W.-J. ZOU, M.-Y. WU, C. SHEN, L. MEI, W.-C. XIONG;
Dept. of Neurosciences, Case Western Reserve Univ., Cleveland, OH

Abstract: Empathy is the ability to attune to and resonate with the emotional states of others, indispensable for social communication. Deficits in empathic responsiveness are a hallmark feature of autism spectrum disorders (ASD). However, the exact mechanisms of empathy are not fully understood. Observational fear, which encompasses the mice (observers) acquiring freezing behavior by observing other mice (demonstrators) subjecting repetitive foot shocks, is a powerful tool for examining affective empathy in rodents. Here, we found that observer male mice (aged 12-16 weeks) exhibited heightened fear responses specifically toward their cagemate siblings and non-littermate cagemates demonstrator male mice, whereas they did not display elevated fear towards strangers or even non-cohabiting sibling mice. These findings illustrate that mice displayed familiarity-dependent observational fear irrespective of their genetic relatedness,

suggesting that factors beyond mere emotional contagion may be involved in the process of observational fear learning. This elevation can be prompted by olfactory signals emanating from stressed co-house conspecifics. This suggests that olfactory cues released by stressed co-house conspecifics may act as mediators in the social transmission of fear in rodents. By contrast, female observer mice showed no discernible difference when exposed to familiar female demonstrator mice. By conducting a thorough brain-wide histological investigation and employing projection-specific optogenetic manipulation, we identified an insular cortex (IC)-basolateral amygdala (BLA) circuit that selectively contributed to the familiarity-dependent observational fear. These findings illustrate that bidirectional manipulation of activity in the IC-BLA pathway plays a critical role in shaping the acquisition of familiarity-dependent observational fear, without affecting the general expression of observational fear. This suggests a potential causal link between IC-BLA activity and familiarity-dependent observational fear. Using a mouse model of mimics a common genetic factor to ASD—a de novo mutation of Cullin 3 (Cul3)—Cul3 mt mice exhibited compromised familiarity-dependent observational fear, characterized by decreased firing activity of IC-BLA neurons and diminished glutamatergic transmission. Together, our study demonstrates that mice exhibited familiarity-dependent observational fear regardless of kinship, highlighting the potential for targeted modulation of specific insular circuits may offer valuable prospects for ASD associated with empathy impairment treatment.

Disclosures: **W. Zou:** None. **M. Wu:** None. **C. Shen:** None. **L. Mei:** None. **W. Xiong:** None.

Poster

PSTR166. Emotion and Social Behavior in Nonhuman Mammals

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR166.04/MM9

Topic: G.04. Emotion

Support: ERC Synergy Grant 'BrainPlay - the self-teaching brain'
Humboldt-Universität zu Berlin
Einstein Center for Neurosciences Berlin

Title: Play and tickling responses map to the lateral columns of the rat periaqueductal gray

Authors: *N. GLOVELI[1,2], J. SIMONNET[1], W. TANG[1], M. CONCHA-MIRANDA[1], E. MAIER[1,3], A. DVORZHAK[4], D. SCHMITZ[4,5,1,2,6], M. BRECHT[1,5,2];
[1]Bernstein Ctr. For Computat. Neuroscinece Berlin, Berlin, Germany; [2]Einstein Ctr. for Neurosciences Berlin, Berlin, Germany; [3]Dept. of Neuropeptide Res. in Psychiatry, Central Inst. of Mental Health, Med. Facul, Univ. of Heidelberg, Mannheim, Germany; [4]Charité Universitätsmedizin Berlin, Berlin, Germany; [5]NeuroCure Cluster of Excellence, Humboldt-Universität zu Berlin, Berlin, Germany; [6]German Ctr. for Neurodegenerative Dis., Berlin, Germany

Abstract: Although play behavior is known to be critical for development, its behavioral implications and underlying neuronal circuits have yet to be fully understood. Behavioral studies have shown that extensive cortical lesions in juvenile rats do not result in a remarkable decrease in social play behavior, leading us to target midbrain regions for their involvement in generating and upholding play. The periaqueductal grey (PAG) is a midbrain structure with a columnar organization surrounding the aqueduct, along the rostral-caudal axis. While the PAG has been linked to a variety of behaviors, including play and laughter, the neuronal basis for these behaviors have not yet been explored. Here, we investigated the involvement of the PAG in play and tickling, combining a behavioral and electrophysiological approach. We studied ticklishness using an interspecific paradigm with the experimenter tickling juvenile rats, utilizing 50 kHz rat vocalizations as a behavioral marker for ticklishness and positive affect. Additionally, we studied chasing hand play behavior and intraspecific social play. Using Neuropixels probes to record PAG neurons in freely moving rats during play and tickling, we found that the PAG responds to play and tickling in a column specific manner. Neurons localized in the lateral column of the PAG are particularly strongly activated, while dorsal columns of the PAG remain largely neutral or inhibited by tickling and play. To assess the functional role of the lateral PAG we performed column specific optogenetic inactivation experiments during tickling and play. Inactivation of the lateral PAG reduces 50 kHz vocalizations during tickling and interferes with social play behavior. We conclude that the lateral columns of the PAG are critically involved in ticklishness and play. These findings point to circuits involving the PAG to be a good candidate for further investigating the neuronal basis of playfulness.

Disclosures: **N. Gloveli:** None. **J. Simonnet:** None. **W. Tang:** None. **M. Concha-Miranda:** None. **E. Maier:** None. **A. Dvorzhak:** None. **D. Schmitz:** None. **M. Brecht:** None.

Poster

PSTR166. Emotion and Social Behavior in Nonhuman Mammals

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR166.05/MM10

Topic: G.04. Emotion

Support: NRF(2021R1A6A3A01087622)
KIST(2E32212)
NRF(RS-2023-00208692)

Title: Neural mechanism underlying modulation of social behavior in response to fearful conditions

Authors: *Y. CHO[1], J. KIM[1,2];
[1]Korea Inst. of Sci. and Technol., Seoul, Korea, Republic of; [2]Div. of Bio-Medical Sci. and Technol., Korea Univ. of Sci. and Technol. (UST), KIST Sch., Seoul, Korea, Republic of

Abstract: Adapting social behavior in response to situational demands is a critical aspect of social functioning in life. How does the brain select appropriate social behaviors in response to various environmental cues? While the prefrontal cortex is the primary neural substrate regulating social behaviors, the extent to which external stimuli can modify social behavior remains largely unknown. Hypothalamus is a key brain region for the integration of sensory information and appropriate motor output to maintain homeostasis. However, the role of hypothalamic circuitry in social adaptation is largely unclear. Here, we established a new behavioral paradigm to observe the change of social behavior by fearful stimuli. It shows that the social interaction was modified after exposure to fearful environment. With this new behavioral paradigm, we also examined the neural activities of hypothalamic circuitry by acquiring calcium activities by utilizing the fiber photometry technique. Hypothalamus shows highly correlative neural signals for social adaptation. Collectively, these results suggest that the hypothalamic circuitry may be a critical modulator for generation of proper social behaviors by fearful conditions.

Disclosures: Y. Cho: None. **J. Kim:** None.

Poster

PSTR166. Emotion and Social Behavior in Nonhuman Mammals

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR166.06/MM11

Topic: G.04. Emotion

Support: F32MH125634
R01MH108665

Title: A Crh+ amygdala cell activity signature for initiation of aggression

Authors: *E. L. NEWMAN[1], K. THREADGILL[1], E. HISEY[1], N. RESSLER-CRAIG[1], H. RIVERA BERMUDEZ[2], K. J. RESSLER[1];
[1]Psychiatry, McLean Hosp., Belmont, MA; [2]Rockefeller Univ., Rockefeller Univ., New York, NY

Abstract: Background: Physical aggression is typically a last resort for settling a dispute; this implies that the transitions between behaviors leading up to an attack represent sequential checkpoints for either escalating or deescalating a confrontation. The likelihood of an aggressive attack increases if the opponent is perceived as threatening; as such, we examine threat-encoding corticotropin-releasing-hormone-expressing central amygdala (*Crh*+ CeA) neurons during the escalation of aggressive behaviors using single-cell calcium imaging, chemogenetics, and localized knockout strategies. **Methods:** Male CRH-ires-Cre mice received intra-CeA Cre-dependent adeno-associated-virus (AAV) to drive expression of the calcium-dependent-fluorescent indicator, GCaMP, in Crh+ CeA cells. Mice were implanted with a gradient index lens over the CeA to record calcium-dependent fluorescence during low- or high-threat

encounters with a submissive or aggressive conspecific, respectively. Additional CRH-ires-Cre males expressed control fluorescent protein, inhibitory Designer Receptors Exclusively Activated by Designer Drugs (iDREADDs) in all Crh+ CeA neurons, or iDREADDs in Crh+ CeA-ventral tegmental area (VTA) projections. DREADD agonist was injected prior to aggression tests to inhibit Crh+ CeA cells. For localized CRH knockout, aggression-naïve or aggression-experienced floxed-Crh mice and wild-type controls received intra-CeA AAV-Cre; two weeks later, mice underwent repeated low-threat aggression testing (2-3 tests/wk for >4 wks). **Results**: Crh+ CeA cells were classified into ensembles during low-threat offensive aggression tests- 41% were active before attack initiation, 32% were active during attacks, and 27% were inactive during aggression. Chemogenetic inhibition of all Crh+ amygdala cells selectively prevented offensive attacks without affecting social approach, contact, or adaptive self-defensive bites toward a high-threat aggressive intruder. Chemogenetically inhibiting Crh+ CeA-VTA projections neurons suppressed - but did not eliminate - offensive attacks. Localized Cre-mediated CRH knockout in the CeA prevented the development of agonistic behaviors in aggression-naïve mice without affecting fighting in experienced aggressors. **Conclusions:** Amygdala CRH signaling is necessary for the initiation of offensive aggression, but not for self-defense, suggesting distinct neural circuitry and signaling substrates for offensive vs. self-defensive aggression. All-optical closed-loop approaches are being used to block attack initiation using Crh+ CeA cell activity to trigger real-time inhibitory optogenetics.

Disclosures: E.L. Newman: None. **K. Threadgill:** None. **E. Hisey:** None. **N. Ressler-Craig:** None. **H. Rivera Bermudez:** None. **K.J. Ressler:** None.

Poster

PSTR166. Emotion and Social Behavior in Nonhuman Mammals

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR166.07/MM12

Topic: G.04. Emotion

Support: Howard Hughes Medical Institute
Salk Institute for Biological Studies
Clayton Foundation
Kavli Foundation
Dolby Family Fund
NIMH R01-MH115920
NIMH R37-MH102441
NCCIH Pioneer Award DP1-AT009925

Title: Acute and chronic social isolation promote diverse behavior repertoires and differentially modify mPFC responses to social contact

Authors: C. R. LEE[1,3], G. P. SCHNEIDER[1,4], A. GARCIA[1,3], T. B. TUAZON[1,3], D. TSIN[1,3], M. G. CHAN[1,4], K. BATRA[1,3], *A. BAKHTI-SUROOSH[2,3], F. H. TASCHBACH[1,3], R.

WICHMANN[1], T. D. PEREIRA[1], M. K. BENNA[3], K. M. TYE[1,3,4,5];
[1]Salk Inst. for Biol. Studies, La Jolla, CA; [2]Salk Inst. for Biol. Studies, San Diego, CA; [3]Univ. of California San Diego, La Jolla, CA; [4]Howard Hughes Med. Inst., La Jolla, CA; [5]Kavli Inst. for Brain and Mind, La Jolla, CA

Abstract: Divergent social behaviors emerge from different durations of social isolation (Lee et al., 2021). However, the exact time course of isolation impacting social behavior and the neural systems and circuits maintaining social homeostasis remain elusive. Previously, we found that dorsal raphe nucleus dopamine neurons mediate a loneliness-like state and innervate the medial prefrontal cortex (mPFC) (Matthews et al., 2016). To explore how the mPFC encodes social information and undergoes a state change following isolation, we used cellular resolution calcium imaging, ultrasonic vocalization (USV) recordings, computer vision, and machine learning tools. To first determine how different durations (2hr, 6hr, 24hr, 7d, 14d, and 28d) of isolation impact social behavior, we performed a juvenile intruder task after isolating adult male mice. We performed pose estimation using SLEAP (Pereira et al., 2022) and a custom pipeline for behavioral feature extraction and unsupervised clustering for behavioral motif discovery. We found an inverse correlation between isolation duration and interaction time (n=108 mice, R^2=0.115, p=0.0023) and that 2 and 6 hours of isolation promotes social interaction with juvenile mice (n=108 mice, F(6,101)=4.715, p=0.0003; GH vs 2hr: p =0.0002; GH vs 6hr: p=0.0409). Despite not detecting differences in interaction time in chronically isolated mice compared to grouphoused mice, unsupervised clustering of behavior features revealed changes in social behavior repertoire, promoting face sniffing and reducing chasing in 7 and 14 day isolated mice (n=54 mice, F(15,192)=2.806, p=0.0006). Next, we found that 2 hours of isolation increased call rate (n=108 mice, F(6,101)=2.831, p=0.0137; GH vs 2hr: p=0.0218) and changed USV acoustic features (n=90 mice, F(5,84)=4.444, p=0.0012; GH vs 2hr: p=0.0352). Finally, we performed calcium imaging using miniature endoscopes in the mPFC of mice engaged in social behavior after group-housing and isolation. We found that isolation (24hr and 7d) increased the responsiveness of mPFC neurons to social contact (n=18 mice, 3938 cells, GH vs 24hr: χ^2 =9.090, p=0.0106; GH vs 7d: χ^2=30.28, p<0.0001) by promoting excitatory responses (n=18 mice, F(2,15)=5.189, p=0.0194; GH vs 24hr: p=0.0548, GH vs 7d: p=0.0146). Additionally, we found an increase in mPFC population trajectory length following isolation compared to a group housed session prior, an effect reversible followed by re-housing (n=6 mice, F(2,6)=16.60, p=0.0036; GHD1 vs 24hr: p=0.0110; 24hr vs GHD2: **p=0.0055). Overall, our findings support a role for mPFC in promoting features of the response to novel social stimuli following social isolation.

Disclosures: C.R. Lee: None. **G.P. Schneider:** None. **A. Garcia:** None. **T.B. Tuazon:** None. **D. Tsin:** None. **M.G. Chan:** None. **K. Batra:** None. **A. Bakhti-Suroosh:** None. **F.H. Taschbach:** None. **R. Wichmann:** None. **T.D. Pereira:** None. **M.K. Benna:** None. **K.M. Tye:** None.

Poster

PSTR166. Emotion and Social Behavior in Nonhuman Mammals

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR166.08/MM13

Topic: H.06. Social Cognition

Support: Mount Sinai BMEII Intramural Grant ANRP
NIH Grant RF1MH117040

Title: The role of macaque frontal face patches in the integration of social and value information

Authors: *C. ELORETTE[1], A. FUJIMOTO[1], S. H. FUJIMOTO[1], L. FLEYSHER[2], B. E. RUSS[4], P. RUDEBECK[3];
[1]Neurosci., Icahn Sch. of Med. at Mount Sinai, New York, NY; [3]Neurosci., [2]Icahn Sch. of Med. At Mount Sinai, New York, NY; [4]Nathan Kline Inst. for Psychiatric Res., Orangeburg, NY

Abstract: Brain regions sensitive to faces have long been observed in both macaques and humans within the temporal and occipital cortex. A subset of these face-sensitive 'patches' have also been identified within prefrontal cortex, but unlike the patches seen in temporal cortex, the specific role of these frontal face patches is still unknown. The most consistently observed frontal face patch, area PO, is located within orbitofrontal cortex (OFC), a region implicated in valuation. In clinical studies, patients with OFC lesions display inappropriate social behavior, suggesting that this area may be critical to combining perceptual representations of faces with internal representations of value, which work in concert to guide social decision making. We hypothesize that area PO, within OFC, specifically acts to update and maintain the value of a face. We used two female rhesus macaques (*Macaca mulatta*) previously trained to perform behavioral tasks during awake functional neuroimaging. We used a behavioral task designed to isolate learned value associations for objects from learned value associations for faces. Animals were trained on different sets of images of macaques faces and objects. They learned to associate a high-value or low-value juice reward flavor with these stimuli. We wanted to test whether area PO was involved in value updating, and if so, if it was specific to value updating for faces. We tested animals in settings where the stimulus-reward associations were periodically reversed, such that a stimulus initially associated with low value reward would become associated with a high value reward and vice versa. Awake whole brain functional images were acquired on a 3T MRI scanner (1.6mm isotropic voxels) while animals performed the reversal task. We examined neural signal changes in the frontal face patches. Our GLM contrast (reversed faces > reversed objects) revealed a unilateral activation in area PO ($p<0.05$) when reward associations were updated for faces, but not for objects. Our results show that Area PO plays a role in updating stimulus-reward associations for faces, as compared to object stimuli. This suggests that this face patch is a critical mediator of face and value information, two streams of information vital for social valuation.

Disclosures: C. Elorette: None. **A. Fujimoto:** None. **S.H. Fujimoto:** None. **L. Fleysher:** None. **B.E. Russ:** None. **P. Rudebeck:** None.

Poster

PSTR166. Emotion and Social Behavior in Nonhuman Mammals

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR166.09/MM14

Topic: H.06. Social Cognition

Support: Canadian Institutes of Health Research (FRN 148365)
BrainsCAN
Natural Sciences and Engineering Research Council of Canada (Discovery grant)

Title: Overlapping temporal networks for face and voice processing in the common marmoset

Authors: *A. DUREUX, A. ZANINI, A. JAFARI, S. EVERLING;
Western Univ., London, ON, Canada

Abstract: Through social communication, various primate species are able to transmit signals among individuals, enabling the expression of intentions and facilitating interaction with other group members, the maintenance of social structure, predator avoidance, and cohesion during daily activities or travel. These signals comprise a range of visual, olfactory, tactile, and auditory cues, with facial expressions and vocalizations serving as the primary modes of communication in the majority of primate species. Our current understanding of the interaction between facial and vocal areas in primate social communication is limited, as the majority of studies have predominantly examined facial and vocal processing in isolation, disregarding the interconnectedness between various vocalizations and corresponding facial expressions. Here, we investigated whether face and vocalization processing are organized in overlapping networks in the temporal face-patches of marmoset cortex, a New World monkey known to have a rich vocal repertoire, and which shares several aspects of pro-social behavior with humans. To achieve this, we employed ultra-high field fMRI conducted in a 9.4T MRI scanner. Our objective was to directly compare the networks involved in face and vocal processing and identify specific brain regions that respond to both faces and vocalizations. For that, we presented to six awake marmosets either marmosets face videos alone, vocalization sounds alone, marmosets face videos accompanied by corresponding vocalizations and the scrambled versions of these three conditions. Our results show that both marmoset vocalizations alone and marmoset faces videos alone, compared to their scrambled versions, activate brain regions associated with facial processing (along the occipitotemporal axis) and vocalization processing (in the primary auditory cortex, cingulate, frontal, and temporal cortices). This suggests the existence of distinct yet partially overlapping networks within the temporal cortex. Through our MVPA searchlight cross-decoding accuracy analysis, we discovered that a subset of patches in temporal cortex responds to both faces and vocalizations, implying that vocalizations are organized in a system similar to the face patch system. Importantly, when face and vocalization stimuli were combined, we observed increased brain activity compared to the sum of responses for each modality, indicating an interaction between the two modalities. These findings offer valuable insights into the integration of faces and vocalizations within the marmoset brain, addressing a previously existing knowledge gap regarding primate social communication.

Disclosures: **A. Dureux:** None. **A. Zanini:** None. **A. Jafari:** None. **S. Everling:** None.

Poster

PSTR166. Emotion and Social Behavior in Nonhuman Mammals

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR166.10/MM15

Topic: H.06. Social Cognition

Support: DFG Emmy Noether Research Group 392443797
NIMH R01MH132236 & BMBF

Title: Testing for grid-like activity in human prefrontal cortex during social navigation

Authors: K. FROLICHS, ***C. KORN**;
Univ. Hosp. Heidelberg, Heidelberg, Germany

Abstract: Humans navigate complex social environments. To do so, they often use personality traits when thinking about themselves and others. Thinking about personality traits reliably and robustly activates medial prefrontal cortex. However, the underlying neural coding principles are unknown. Here, we tested whether coding principles that have been described for navigation in physical (Doeller et al., 2010; Bellmund et al., 2018) and conceptual spaces (Constantinescu et al., 2016; Schafer & Schiller, 2018) play a similar role for navigating a "personality trait space." That is, we tested whether a grid-like, hexagonal activity pattern—which has been identified in the medial prefrontal cortex for moving in physical and conceptual spaces—relates to navigating a personality trait space. In a preregistered study (https://osf.io/pxs7m), we collected behavioral and fMRI data on two consecutive days. In the scanner, participants (total n=42; final n=36) saw moving bars that corresponded to ratings on two orthogonal personality traits (diligent and generous). In a series of behavioral tasks, we found that participants learned the positions of six individuals in this two-dimensional space, without being aware that these six individuals could be located in such a space. Participants memorized these learned positions and were able to generalize them to related personality traits. Furthermore, participants could use this personality trait space to position their friends. However, we have not found evidence for grid-like activity in the medial prefrontal cortex by using the preregistered fMRI analyses. These analyses included whole-barain maps and ROI analyses. We will discuss reasons why results from previous tasks may not generalize to our task.

Disclosures: K. Frolichs: None. **C. Korn:** None.

Poster

PSTR166. Emotion and Social Behavior in Nonhuman Mammals

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR166.11/MM16

Topic: A.09. Adolescent Development

Support: NSF #1926818

Title: Multi-animal pose estimation in prairie voles during social behavior using SLEAP

Authors: N. MILMAN[1], L. BUENO-JUNIOR[2], *C. TINSLEY[3], J. LOEUNG[4], B. WATSON[2], M. LIM[5];
[1]Oregon Hlth. & Sci. Univ. Behavioral Neurosci., Portland, OR; [2]Univ. of Michigan, Ann Arbor, MI; [3]Oregon Hlth. and Sci. Univ., Portland, OR; [4]VA Portland Hlth. Care Syst., Portland, OR; [5]Veterans Affairs Portland Hlth. Care Syst., Portland, OR

Abstract: Background: Prairie voles (*Microtus ochrogaster)* are a highly social, wild rodent species that display a variety of affiliative social behaviors toward familiar conspecifics, including prolonged huddling behavior. Humans display a spectrum of affiliative social behaviors ranging from explicit to more subtle features. Due to the nature of manually scored behavior and intrinsic features of prairie voles, the field has been limited in our characterization of subtle features of affiliative social behavior. Advancements in user-friendly tools to estimate animal behavior including Social Leap Estimates Animal Pose (SLEAP) has enabled accurate and precise pose-estimation of two freely-behaving prairie voles during social interactions.
Methods: Adolescent (P28) same-sex sibling cagemates were placed in a novel-home cage to explore and interact. Animal pose was annotated and trained in SLEAP to generate pose-estimation for both animals across all videos. Coordinates were transformed in MATLAB to derive subtle features of affiliation including body-direction and the temporal sequence of physical interactions between siblings.
Results: A SLEAP model trained on 324 frames across forty unique social investigation videos, had precision of 0.98 and recall of 0.93 in identifying vole nose, ears, back shoulder and tail base. In this model, the average distance between ground truth and prediction was 16.25 pixels (about 3% of a vole's body length), with distance for $50^{th}/75^{th}/90^{th}$ percentile = 6.89/13.69/44.54 pixels.
Conclusions: Our data suggests that multi-animal pose-estimation of freely behaving prairie voles is feasible. Transformation of pose-estimates enables investigation of subtle features of affiliative behavior inaccessible to manual scoring approaches. Application of SLEAP in a diversity of social behavior paradigms will improve translation and reproducibility of animal research.

Disclosures: N. Milman: None. **L. Bueno-Junior:** None. **C. Tinsley:** None. **J. Loeung:** None. **B. Watson:** None. **M. Lim:** None.

Poster

PSTR167. Cocaine: Cell Signaling, Circuitry, and Neurophysiology

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR167.01/MM17

Topic: G.09. Drugs of Abuse and Addiction

Support: ANR #21-CE16-0002-01
BBRF #300015

Title: Electrophysiological analysis of intrinsic and synaptic properties of subthalamic neurons in a new rodent model of cocaine addiction

Authors: M. TAPIA[1], M. WILLIAMS[1], A. AWADA[1], L. VIGNAL[1], **C. BAUNEZ**[1], J.-M. GOAILLARD[2], *M. DEGOULET[1];
[1]INT CNRS & Aix-Marseille Univ., Marseille, France; [2]INT INSERM & Aix-Marseille Univ., Marseille, France

Abstract: The subthalamic nucleus (STN) plays a key role in the cortico-basal ganglia-thalamo-cortical circuits. Besides its well-known role in motor functions, preclinical studies indicate that the STN is an important hub for controlling cocaine seeking and taking behaviors. In particular, an aberrant increase of STN low frequencies can predict compulsive-like cocaine seeking behavior in the rat. Because of the intrinsic rhythmicity abilities of STN neurons, such oscillatory changes could arise from modifications in the properties of synaptic inputs or in intrinsic properties. However, the cellular and molecular mechanisms underlying STN abnormal oscillatory activity have yet to be studied. Here, we performed patch-clamp recordings on STN neurons of adult male rats previously subjected to an 'Intra-Escalation Punishment' procedure (8h/d, 5 or 15d), consisting in a 2h cocaine seeking phase, in which seeking responses are randomly punished with a mild electric foot shock, inserted between two 3h phases of escalation, during which they can freely self-administer cocaine. This procedure allows the concomitant development of loss of control over cocaine intake and compulsive-like seeking behavior. To better characterize the electrical signature of this neuronal population, we performed recordings of STN neurons (n=22, 14 animals) in *ex vivo* horizontal brain slices, using current- and voltage-clamp protocols to investigate i) intrinsic excitability properties and ii) internal capsule-evoked excitatory synaptic responses. We first focused on the properties of evoked AMPA and NMDA currents. Global analysis revealed no significant difference in the amplitude of the glutamatergic response and no significant modification of the AMPA/NMDA ratio, even though both parameters displayed significant variability. Current-clamp recordings, currently under analysis, may however reveal changes in neurons intrinsic properties.These preliminary results suggest that the Hebbian plasticity capacity of STN glutamatergic inputs is not modified in this model of cocaine addiction. However, further analysis taking into account rats' compulsive phenotype, and comparison with acute cocaine models may help us to understand the different molecular mechanisms underlying cocaine addiction.

Disclosures: **M. Tapia:** None. **M. Williams:** None. **A. Awada:** None. **L. Vignal:** None. **C. Baunez:** None. **J. Goaillard:** None. **M. Degoulet:** None.

Poster

PSTR167. Cocaine: Cell Signaling, Circuitry, and Neurophysiology

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR167.02/MM18

Topic: G.09. Drugs of Abuse and Addiction

Support: DA031900
NSF GRFP

Title: Optostimulation of ventral tegmental area dopamine neurons increases cocaine potency at dopamine transporters

Authors: *A. R. BASHFORD[1], P. J. CLARK[2], R. A. ESPAÑA[2];
[1]Drexel Univ. Col. of Med. Neurosci. Program, Philadelphia, PA; [2]Neurobio. and Anat., Drexel Univ. Col. of Med., Philadelphia, PA

Abstract: Accumulating evidence suggests that greater dopamine transporter (DAT) sensitivity to cocaine is associated with escalation of cocaine taking and exaggerated motivation for cocaine. Across several experiments, we have shown that pharmacological manipulations known to alter ventral tegmental area (VTA) dopamine neuron activity result in rapid changes in DAT sensitivity to cocaine in the nucleus accumbens, suggesting that dopamine cell body activity can regulate DAT function at distal sites. Consistent with this hypothesis, we recently demonstrated that chemogenetic activation of VTA dopamine neurons leads to increased DAT sensitivity to cocaine, further suggesting the likelihood that dopamine neuron activity influences DAT function. Despite this evidence, all of our previous studies employed G-protein coupled receptor (GPCR) manipulations and thus, it is unclear whether changes in DAT function are mediated through changes in DA neuron activity or GPCR signaling cascades. To initially address this issue, we used optogenetics to activate VTA dopamine neurons in vivo and measured resulting changes in dopamine transmission in brain slices containing the nucleus accumbens core. Tyrosine hydroxylase Cre rats received bilateral injections of channelrhodopsin in the VTA and after three weeks of virus incubation, received optostimulation of VTA dopamine neurons. After thirty minutes of stimulation, we performed fast-scan cyclic voltammetry to measure dopamine release and uptake at baseline, as well as DAT sensitivity to cocaine. We found that activation of VTA dopamine neurons did not affect baseline release or uptake, similar to what we observed with chemogenetic manipulations. However, we did observe that activation of dopamine neurons increased DAT sensitivity to cocaine. Although the mechanisms through which dopamine neuron activity influences DAT function remain unclear, these results suggest that changes in DAT sensitivity to cocaine are not dependent on GPCR signaling cascades. Consequently, these findings further support the tenet that simply driving the activity of VTA dopamine neurons is sufficient to alter DAT function. Future work will be needed to explore how specific changes in DA neuron activity (e.g., firing rate, firing patterns, membrane potential) drive alterations in DAT cocaine sensitivity, and to what extent changes in dopamine neuron activity influence cocaine-associated behavior.

Disclosures: A.R. Bashford: None. **P.J. Clark:** None. **R.A. España:** None.

Poster

PSTR167. Cocaine: Cell Signaling, Circuitry, and Neurophysiology

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR167.03/MM19

Topic: G.09. Drugs of Abuse and Addiction

Support: NIDA 1U18DA052497
NIDA R01DA048153
NINDS F99NS129175

Title: Regulator of G protein Signaling-12 modulates cocaine withdrawal-associated behavior and cocaine intravenous self-administration in C57BL/6J mice

Authors: *A. N. WHITE[1,2,3], B. J. MENARCHEK[1,2,3], D. P. SIDEROVSKI[5], V. SETOLA[2,3,4]; [1]Physiology, Pharmacology, and Toxicology, [2]Neurosci., [3]Rockefeller Neurosci. Inst., [4]Behavioral Med. and Psychiatry, West Virginia Univ., Morgantown, WV; [5]Pharmacol. and Neurosci., Univ. of North Texas Hlth. Sci. Ctr., Fort Worth, TX

Abstract: Stimulant misuse is a pressing public health issue, as stimulant-use-associated deaths have increased over the past decade. Given the lack of available treatments for conditions such as cocaine use disorder (CUD), further investigations of the processes modulating stimulant reward and of potential therapeutic targets are necessary. Our group discovered that mice lacking Regulator of G protein Signaling-12 (RGS12) are insensitive to the locomotor-stimulating effects of low-to-moderate doses of cocaine. In the ventral striatum, RGS12-null mice exhibit 1) increased protein levels and/or uptake activity for the major molecular targets of cocaine (*i.e.*, the dopamine and serotonin transporters DAT and SERT) and 2) reduced electrically stimulated dopamine release in the nucleus accumbens core. Given the influence of ventral striatal DAT, SERT, and extracellular dopamine on the pleiotropic CNS effects of cocaine, our group is now investigating RGS12 as a potential CUD drug target. To validate RGS12 as a potential target for CUD, cocaine or vehicle was administered to wild-type and RGS12-null C57BL/6J mice for 12 days (*t.i.d.*, 5-20 mg/kg per injection), and behaviors associated with discontinuation of subchronic cocaine administration were recorded following the last injection. Wild-type mice displayed a cocaine-discontinuation-associated increase in grooming compared with saline-treated mice. In contrast, both saline- and cocaine-treated RGS12-null mice displayed no cocaine-discontinuation-associated alterations in grooming behavior, suggesting that loss of RGS12 function (by either genetic or pharmacological manipulation) may have desirable effects on at least some aspects of cocaine withdrawal. Cocaine intravenous self-administration (IVSA) studies were then conducted to evaluate acquisition and extinction of volitional cocaine intake in wild-type and RGS12-null mice. While the rates of acquisition of cocaine IVSA were similar in wild-type and RGS12-null mice, RGS12-null mice administered more infusions of cocaine (0.5 mg/kg/infusion) than did wild-type mice. During the extinction phase of cocaine IVSA, wild-type mice maintained elevated active lever responses for the 10-day extinction period, while RGS12-null mice exhibited significantly reduced active lever responses. These data, along with observation that RGS12 loss affects IVSA dose-response and breakpoint, suggest that RGS12 modulation has a potential as a CUD therapy. Future studies will evaluate the effect of RGS12 on motivation for cocaine reward and examine RGS12 as a druggable target for CUD.

Disclosures: A.N. White: None. **B.J. Menarchek:** None. **D.P. Siderovski:** None. **V. Setola:** None.

Poster

PSTR167. Cocaine: Cell Signaling, Circuitry, and Neurophysiology

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR167.04/MM20

Topic: G.09. Drugs of Abuse and Addiction

Title: Epigenetic regulation of drug addiction susceptibility by Nr4a1 enhancer variant

Authors: *J. J. WINTER[1], M. T. WOOLF[1], K. S. KRICK[1], S. ZHANG[1], E. A. HELLER[2]; [2]Dept. of Systems Pharmacol. and Translational Therapeut., [1]Univ. of Pennsylvania, Philadelphia, PA

Abstract: Despite the high numbers of cocaine dependent individuals in the US, effective and targeted therapies are still missing. Our research focuses on persistent cocaine-induced transcriptional changes in the brain during abstinence. We were able to identify the nuclear orphan receptor Nr4a1 and its downstream target cocaine and amphetamine regulated transcript (Cartpt) as critical players during abstinence. Nr4a1 is a transcription factor acting as an immediate early gene upon various stimuli. We could show that cocaine regulates Nr4a1 and Cartpt expression via histone post-translational modifications (HPTMs). In addition, we could show that CRISPR-mediated regulation of Nr4a1 bidirectionally modulates cocaine behavior. We aim to further characterize this potential master regulator of cocaine-induced transcription during abstinence to enable the development of new therapeutics for cocaine dependence. We also aim to further characterize the role of Cartpt during late abstinence. Recent evidence shows that HPTMs regulate gene expression via chromatin accessibility and transcription factor recruitment. To investigate the functional relevance of certain HPTMs, we make use of a novel method called ICuRuS currently established by our group to perform transcriptional and epigenetic profiling of specific neuronal cell types in the mouse brain. We also apply epigenetic editing using a modified dCas9 protein to further functionally characterize HPTMs identified by ICuRuS. In a second approach we use chromatin conformation capture and a CRISPR-dCas9-based approach to identify and characterize potential enhancer regions of Nr4a1 and Cartpt affected by cocaine abuse. Since genetic variation plays a major role in addiction neurobiology, we are screening these cis-regulatory elements for potential genetic variants to validate addiction-associated risk loci previously identified in genome-wide association studies.

Disclosures: **J.J. Winter:** None. **M.T. Woolf:** None. **K.S. Krick:** None. **S. Zhang:** None. **E.A. Heller:** None.

Poster

PSTR167. Cocaine: Cell Signaling, Circuitry, and Neurophysiology

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR167.05/MM21

Topic: G.09. Drugs of Abuse and Addiction

Support: Davidson Foil Family Research Award

Title: Investigating the molecular mechanisms of cocaine action using Caenorhabditis elegans

Authors: *E. WILLIAMS;
Davidson Col., Davidson, NC

Abstract: Thousands of people in the US abuse cocaine, often with consequences such as toxicity and addiction. Cocaine's mode of action is incompletely understood, resulting in a lack of pharmaceutical treatment. Using the *C. elegans* egg laying circuit as a genetically tractable, relatively simple neural circuit that uses conserved neurotransmitter systems, we showed that acute treatment with cocaine stimulates a quantifiable egg laying behavior and that acetylcholine signaling and release genes are directly required for this behavior. We also showed that mutants lacking *mod-1*, a serotonin-gated chloride channel, show an increase in egg output compared to wild type worms but do not observe any effect in other serotonin signaling mutants we tested. Recently, we showed that the increased egg laying we observed in the *mod-1* null is fully suppressed by a null allele of the single *C. elegans* tryptophan hydroxylase, *tph-1(n4622)*, suggesting that the increased egg laying observed in *mod-1* is serotonin dependent. To understand the downstream effects of cocaine stimulation in neurons, we separated different populations of neurons from cocaine-treated and controlled animals by FACS sorting and identified differentially expressed genes. Our data show that most gene expression differences are neuron-specific, but some genes are differentially expressed in the same way in all neurons tested. We are currently investigating the top candidate genes from our RNASeq experiment, starting with conserved genes exhibiting high fold change, significant differential expression and prioritizing genes with reported function in neuroplasticity or with previously demonstrated roles in substance use disorders.

Disclosures: E. Williams: None.

Poster

PSTR167. Cocaine: Cell Signaling, Circuitry, and Neurophysiology

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR167.06/MM22

Topic: G.09. Drugs of Abuse and Addiction

Support: NIH Grant R00DA042934

Title: Cocaine-induced alterations in the nucleus accumbens and dorsolateral striatum to reward predictive cues following outcome devaluation

Authors: *C. CORBETT, L. ADRIAN, T. J. SLOAND, M. NIEDRINGHAUS, E. WEST; Cell Biol. and Neurosci., Rowan Univ., Stratford, NJ

Abstract: Substance use disorders (SUDs) are characterized by a continuation of maladaptive behavior despite negative consequences. Characterizing the underlying processes contributing to the ability to change or stop behavior in response to updated expected outcomes is critical for understanding the neurobiological alterations in SUDs. We used *in vivo* electrophysiology to investigate how cocaine exposure leads to aberrant patterns of neural activity in the nucleus accumbens (NAc) and dorsolateral striatum (DLS) to reward predictive cues following outcome devaluation. Mildly water-regulated Long-Evans rats underwent self-administration for cocaine or water and saline (i.v.) (2 h/d for 10 days). After 3 weeks of abstinence, rats were presented with two distinct cues as conditioned stimuli (CS+; predicting a sucrose or grain pellet) and two cues that did not predict a reward for 10 days. Following conditioning, rats underwent a conditioned taste aversion (LiCl, i.p.) to the sucrose pellets. Then, rats were tested on the same Pavlovian task (under extinction) to evaluate their ability to avoid the CS+ associated with the devalued outcome (i.e., sucrose pellets). Electrophysiological recordings showed neuronal populations that were phasic to CS+ [excited or inhibited] or nonphasic (no response). In the NAc, control rats showed a greater % of phasic neurons to NonDevalued CS+ compared to the Devalued CS+, while cocaine rats showed similar phasic responses to both cues. There was a greater % of NAc neurons that selectively responded to the NonDevalued CS+ compared to Devalued CS+ in control rats (ND:61%, both:22%, D: 15%). In cocaine rats, a similar % of NAc neurons were selective to the NonDevalued and Devalued CS+ (ND:39%, both:22%, D:39%). In the DLS, control rats showed a greater % of phasic neurons to NonDevalued CS+ compared to the Devalued CS+, while cocaine rats showed similar phasic responses to both cues. There was a greater % of DLS neurons that selectively responded to the NonDevalued CS+ compared to Devalued CS+ in control rats, and few neurons that responded to both cues (ND: 73%, both: 8%, D: 18%). However, in cocaine rats, a greater % of neurons were selective to the Devalued CS+, and most neurons responded to both cues (ND: 16%. both: 53%, D:31%). Together these findings suggest that the altered neural encoding in cocaine-exposed rats in the nucleus accumbens and dorsolateral striatum may contribute to impaired ability to stop avoiding a cue paired with a devalued outcome.

Disclosures: C. Corbett: None. **L. Adrian:** None. **T.J. Sloand:** None. **M. Niedringhaus:** None. **E. West:** None.

Poster

PSTR167. Cocaine: Cell Signaling, Circuitry, and Neurophysiology

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR167.07/MM23

Topic: G.09. Drugs of Abuse and Addiction

Support: NIH Grant DA046794

Title: SSRI potentiation of methylphenidate-induced gene regulation in the striatum: effects of SSRI vilazodone are tempered by 5-HT1A receptor

Authors: *H. STEINER, C. MOON, **M. HRABAK**;
ROSALIND FRANKLIN UNIV OF MED AND SCIENCE, NORTH CHICAGO, IL

Abstract: Selective serotonin reuptake inhibitors (SSRIs), including fluoxetine, are frequently combined with medical psychostimulants such as methylphenidate (Ritalin), for example, in the treatment of attention-deficit hyperactivity disorder/depression comorbidity. Co-exposure to these medications also occurs with misuse of methylphenidate as a recreational drug by patients on SSRIs. Methylphenidate, a dopamine reuptake blocker, produces moderate addiction-related gene regulation. Findings show that SSRIs such as fluoxetine when given together with methylphenidate potentiate methylphenidate-induced gene regulation in the striatum, mimicking cocaine effects, thus confirming a facilitatory action of serotonin on addiction-related processes. These SSRIs may thus increase methylphenidate's addiction liability. Here, we investigated the effects of a novel SSRI, vilazodone, on methylphenidate-induced gene regulation. Vilazodone differs from prototypical SSRIs in that, in addition to blocking serotonin reuptake, it has 5-HT1A partial agonist properties. Studies showed that stimulation of the 5-HT1A serotonin receptor tempers serotonin input to the striatum. We compared the effects of vilazodone with those of fluoxetine on striatal gene regulation (zif268, substance P, enkephalin) by in situ hybridization histochemistry. Our results show that acute treatment with vilazodone (10-20 mg/kg) potentiated locomotor activity induced by methylphenidate (5 mg/kg). However, in contrast to fluoxetine, vilazodone did not potentiate methylphenidate-induced gene regulation in the striatum. Moreover, blocking 5-HT1A receptors by the selective 5-HT1A receptor antagonist WAY-100635 (0.5 mg/kg) unmasked a potentiating effect of vilazodone on methylphenidate-induced gene regulation, thus demonstrating an inhibitory role for 5-HT1A receptors in gene regulation. Our findings suggest that vilazodone may serve as an adjunct SSRI with diminished addiction facilitating properties and identify the 5-HT1A receptor as a potential therapeutic target to reduce the addiction risk.

Disclosures: H. Steiner: None. **C. Moon:** None. **M. Hrabak:** None.

Poster

PSTR167. Cocaine: Cell Signaling, Circuitry, and Neurophysiology

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR167.08/MM24

Topic: G.09. Drugs of Abuse and Addiction

Support: Grant BES-2016-076353 funded by MCIN/AEI/10.13039/501100011033 and by "ESF Investing in your future"
Plan Nacional de Drogas 2017: PND-132400
Grant PGC2018-095980-B-100 funded by MCIN/AEI/10.13039/501100011033 and por "ERDF A way of making

Europe"
AICO 2021/215 funded by Conselleria d'Innovació, Universitats, Ciència i Societat Digital and by "ESF Investing in your future"
UJI-B2020-1 funded by Plan de Promoción de la Investigación

Title: Effect of cerebellar perineuronal nets removal in lobule VII on addiction circuit activity

Authors: *A. SANCHEZ-HERNANDEZ[1], A. FÁBREGA-LEAL[1], P. IBÁÑEZ-MARÍN[1], E. MARÍN-SAMPIETRO[1], O. RODRÍGUEZ-BORILLO[1], L. ROSELLÓ-JIMÉNEZ[1], L. FONT[1], M. SOLINAS[2], M. MIQUEL[1];
[1]Basic and Clin. Psychology and Psychobiology, Univ. Jaume I, Castelló de la Plana, Spain; [2]INSERM U1084/University of Poitiers, Poitiers, France

Abstract: Growing evidence has revealed the new roles of the cerebellum in brain functions impaired in drug addiction. Moreover, recent research has shown that the cerebellum modulates the activity of the canonical addiction circuit. Addictive drugs induce aberrant activation of plasticity mechanisms that underlie learning and memory, including those that restrict synaptic modifications to stabilize memory. Perineuronal nets (PNNs) are specializations of the extracellular matrix that enwraps the soma and proximal dendrites of some subsets of neurons and have been linked to synaptic stabilization in drug-induced memory. In the cerebellar cortex, the only neurons that express PNNs are Golgi interneurons and Lugaro cells. As inhibitory interneurons, the primary role of Golgi cells is to control and synchronize the glutamatergic activity of granule cells and mossy fibers to regulate Purkinje neuronal output. A previous investigation from our lab showed a dynamic regulation of PNN expression after extended cocaine self-administration. PNN expression around Golgi interneurons decreased 24 hours after the last session of cocaine self-administration but increased after protracted abstinence. The present study aimed to investigate the effects of PNN removal from lobule VII (LVII) of the vermis on the incubation of drug-seeking. We also explored the impact of PNN disruption on neural activity in some relevant areas of the addiction circuit. We assessed the effect of PNN digestion using the bacterial enzyme Chondroitinase ABC (ChABC) after rats were allowed to self-administer cocaine during an extended (6h) or restricted (1h) access for 12 days. The expression of Golgi-bearing PNNs and C-Fos in the granule cell layer of LVII increase after protracted abstinence. We then explored the effects of PNN disruption following 24 hours or 28 days from the last self-administration session. We found a faster decline in drug-seeking after PNN disruption in LVII. PNN removal prevented increased C-Fos expression in the IL after extended access to cocaine self-administration. These findings involve lobule VII of the vermis in drug-seeking and suggest that PNNs play a key role in the stabilization of plasticity modifications underlying the incubation of drug-seeking during protracted abstinence. Finally, they indicate that cerebellar PNNs might have a role in regulating distal regions such as the IL cortex.

Disclosures: A. Sanchez-Hernandez: None. **A. Fábrega-Leal:** None. **P. Ibáñez-Marín:** None. **E. Marín-Sampietro:** None. **O. Rodríguez-Borillo:** None. **L. Roselló-Jiménez:** None. **L. Font:** None. **M. Solinas:** None. **M. Miquel:** None.

Poster

PSTR167. Cocaine: Cell Signaling, Circuitry, and Neurophysiology

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR167.09/MM25

Topic: G.09. Drugs of Abuse and Addiction

Support: K01DA043615
R01DA041528

Title: Cortical activity underlying motivated attention to salient cues in individuals with cocaine use disorder

Authors: *T. BEL-BAHAR, R. Z. GOLDSTEIN, M. A. PARVAZ;
Psychiatry, Icahn Sch. of Med. at Mount Sinai, New york, NY

Abstract: Heightened motivated attention to drug-related over other salient cues is a hallmark of substance use disorder and greater relapse risk. While brain networks involved with motivated attention to cues (or cue-reactivity) have been detailed with high spatial resolution using fMRI, the low temporal resolution of fMRI limits the examination of cue-reactivity at a sub-second level. EEG-derived event-related potentials, such as the late positive potential (LPP), have been used to reliably index motivated attention to cues, including drug cues in people with substance use disorders, at this higher temporal resolution. However, gaps in current knowledge exist about the cortical generators of drug cue-induced LPPs. EEG data was collected from 86 individuals with cocaine use disorder (≤ 30 days abstinent) while viewing pleasant, unpleasant, neutral, and cocaine-related pictures (LPP-generating task). We processed data into single-subject -500 to 2000 ms trial averages for each condition. Event-related sensor and source estimates were examined separately for each of the three affective conditions (i.e., pleasant, unpleasant, and drug) contrasted against the neutral condition. Within-subjects permutation t-tests for salience effects were performed for early (400-1000 ms) and late (1000-2000 ms) LPP time windows. Across all participants and only for the early time window, the LPP amplitude was higher at fronto-central sensors in the unpleasant compared to neutral condition ($pFDR<0.0008$). On the source level, the drug versus neutral contrast yielded greater activity in the left cingulate, parahippocampal gyrus, precentral, superior parietal, and right paracentral, posterior cingulate, caudal midfrontal, and posterior superior temporal sulcus ($pFDR<0.0007$). These EEG-derived source maps of drug cue-reactivity overlap with brain regions identified with fMRI-assessed cue-reactivity in emotion regulation, extending current knowledge about cortical generators of the LPP elicited by salient drug relative to neutral cues in substance use disorder. These preliminary results pave the way for identifying temporally and spatially specific mechanisms of motivated attention to salient cues in substance use disorder and point to the utility of EEG brain imaging for precision psychiatry.

Disclosures: **T. Bel-Bahar:** A. Employment/Salary (full or part-time); Icahn School of Medicine at Mount Sinai. **R.Z. Goldstein:** A. Employment/Salary (full or part-time); Icahn School of Medicine at Mount Sinai. **M.A. Parvaz:** A. Employment/Salary (full or part-time); Icahn School of Medicine at Mount Sinai.

Poster

PSTR167. Cocaine: Cell Signaling, Circuitry, and Neurophysiology

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR167.10/NN1

Topic: G.09. Drugs of Abuse and Addiction

Support: Austrian Science Fund (FWF) P27852-B21, T758-BBL

Title: Protective effect of social interaction when available as an alternative to cocaine: involvement of protein kinases in the nucleus accumbens shell

Authors: *I. M. AMARAL, C. LEMOS, A. SALTI, A. HOFER, R. EL RAWAS;
Med. Univ. Innsbruck, Innsbruck, Austria

Abstract: Social interaction, when available in a distinct context from the one associated with drug consumption, is able to eliminate preference for cocaine and to prevent against cocaine relapse. However, it is still unclear how this protecting effect is mediated. We examined the involvement of nucleus accumbens (NAc) calcium/calmodulin-dependent protein kinase II (CaMKII) (experiment A), as well as the corticotropin-releasing factor (CRF) system (experiment B), in the expression of reward-related learning of cocaine *vs* social interaction reward. In the first experiment (A), male Sprague Dawley rats (8w old) were subjected to a social interaction or cocaine (15 mg/kg i.p.) conditioned place preference (CPP) protocol. A saline CPP group was used as control. We observed that rats that expressed social interaction CPP had increased NAc αCaMKII levels, when compared to control rats. Moreover, a bilateral intra-NAc core (NAcC) or shell (NAcS) infusion of the CaMKII inhibitor KN-93 (6 µg/0.5 µL/side) was performed before testing the rats in a concurrent CPP, where social interaction was available as an alternative to the drug. Whereas vehicle infusions led to equal preference for both stimuli, inhibition of CaMKII in the NAcS, but not in the NAcC, shifted the rats' preference toward the cocaine-associated context. These results suggest that social interaction reward engages NAcS CaMKII. In a second experiment (B), rats underwent cocaine or social interaction CPP. Firstly, we evaluated stress markers and found that social interaction CPP decreased the percentage of incorrect cephalocaudal grooming transitions (behavioral marker) as well as corticosterone, to the level of naïve untreated rats. Furthermore, in another set of experiments, rats were infused (i.c.v.) with either CRF (1 µg/5µl) or CRF receptor antagonist α-helical CRF (10 µg/5µl), 1 hour prior to conditioning for cocaine CPP. CRF-infused rats displayed increased preference for cocaine - an effect reversed in rats that were infused with α-helical CRF. Importantly, in another group of rats that underwent concurrent CPP, the previously observed CRF-induced increase in cocaine preference was completely reversed to the level of α-helical CRF-infused rats. This effect was paralleled by a decrease in the percentage of cephalocaudal grooming transitions and p38 MAPK expression (stress marker) in the NAcS. These findings suggest that social interaction positive effects are mediated through a decrease in stress levels. We propose that the beneficial effects of social interaction in the NAcS are dichotomized into rewarding effects mediated via CaMKII and anti-stress effects mediated via P38 MAPK.

Disclosures: **I.M. Amaral:** None. **C. Lemos:** None. **A. Salti:** None. **A. Hofer:** None. **R. El Rawas:** None.

Poster

PSTR167. Cocaine: Cell Signaling, Circuitry, and Neurophysiology

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR167.11/NN2

Topic: G.09. Drugs of Abuse and Addiction

Support: NIIMES UNIVERSITY

Title: Higher motivation for food than cocaine correlate low cAMP, changes in neuron structure and specific biomarkers in the absence of 5-HT4 receptors

Authors: *V. COMPAN[1], S. CHOQUART[2];
[1]Nimes Univ., Nimes, France; [2]SCIENCES, BRAINS LABORATORY S.A.S., NIMES, France

Abstract: In cells of the nucleus accumbens, activation of a cAMP signaling pathway is a means of transforming an immediate reduction of drugs' rewarding effect into a durable dependence. After recruiting cAMP-response element binding protein (CREB)-binding protein, the resultant phosphorylated CREB (pCREB) favors the expression of some genes (FosB, ΔFosB) to the detriment of others (methyltransferase G9a of histone), from where come changes in neuron morphology. Serotonin (5-HT, 5-hydroxytryptamine) volume transmission trough different receptors acts on cAMP signaling and modulates the activity of the reward neural pathways. Here, we examine how the absence of one of the 5-HT receptor subtypes, the Gs-coupled serotonin 4 receptors (5-HT_4Rs), impacts morpho-functional effects of cocaine. Cocaine failed to increase the levels of both cAMP and pCREB in the accumbens in the 5-HT_4R knockout (KO) mice. The resultant expression of FosB and ΔFosB was attenuated. Under basal conditions, the mRNA levels of the G9a in the accumbens were higher in mutants than wild-type animals. A reduced number of dendritic spines in the accumbens was also observed in the mutants. Mutants are less motivated to self-administer cocaine but more motivated to consume food following chronic restriction. Hence, high vulnerability to overeating and low cocaine dependence are associated with low cAMP-dependent pathway activity and reduced numbers of dendritic spines in the nucleus accumbens in the absence of 5-HT_4 receptors.

Disclosures: **V. Compan:** None. **S. Choquart:** None.

Poster

PSTR167. Cocaine: Cell Signaling, Circuitry, and Neurophysiology

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR167.12/NN3

Topic: G.09. Drugs of Abuse and Addiction

Support: NIH Grant R21DA0466000
NIH Grant T32NS115664

Title: Glutamate dynamics of the tripartite synapse in the dorsolateral striatum of cocaine-seeking rats

Authors: *K. VEROS, P. WEST, K. WILCOX, K. KEEFE;
Univ. of Utah, Salt Lake City, UT

Abstract: The transition between goal-directed and habitual control over drug-related behaviors is thought to facilitate the shift from drug abuse to drug addiction. The formation of automatic, habitual behaviors is mediated by potentiated glutamate signaling and synaptic strengthening of medium spiny neurons (MSNs) in the dorsolateral striatum (DLS), though DLS glutamate dynamics and synaptic plasticity mechanisms have yet to be fully characterized in the context of habitual drug-seeking behavior. Prolonged cocaine experience and withdrawal has been previously associated with decreased expression of astrocyte-specific glutamate transporters as well as enduring changes in synaptic strength of MSNs in the nucleus accumbens, each of which are considered to promote relapse behaviors. Additional evidence from cocaine-experienced rats indicates the dorsal striatum may exhibit similar alterations to glutamate transporters and glutamate signaling, but these changes remain to be determined in association with goal-directed or habitual control over drug-seeking behavior. Therefore, we used a chained seeking-taking cocaine self-administration paradigm to classify rats as goal-directed or habitual in their cocaine-seeking based on sensitivity to outcome devaluation. Assessments of glutamate clearance via whole-cell patch-clamp recordings of synaptic transporter currents (STCs) in astrocytes of the DLS following a single stimulation or high frequency stimulation revealed no differences in glutamate clearance in cocaine-experienced rats classified as goal-directed or habitual in their cocaine-seeking in relation to yoked-saline control. Western blot analysis of glutamate transporter-1 (GLT-1) protein in the DLS further revealed no differences in GLT-1 protein expression in cocaine-experienced rats of all behavioral classifications. Characterization of synaptic strength via whole-cell patch-clamp recordings of evoked excitatory postsynaptic currents (EPSCs) in MSNs of the DLS at a range of holding potentials provided insight to postsynaptic plasticity mechanisms involving AMPA and NMDA receptors. While no significant differences were observed between averaged MSN responses, variability of AMPA to NMDA EPSC ratios in MSNs from cocaine-experienced rats suggests synaptic changes may occur with cell-type-specificity that requires further investigation. Together, these results suggest DLS glutamate dynamics of the tripartite synapse are largely unaltered by cocaine experience, regardless of whether cocaine-seeking behavior is under goal-directed or habitual control.

Disclosures: K. Veros: None. **P. West:** None. **K. Wilcox:** None. **K. Keefe:** None.

Poster

PSTR167. Cocaine: Cell Signaling, Circuitry, and Neurophysiology

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR167.13/NN4

Topic: G.09. Drugs of Abuse and Addiction

Support: NIH R01 Grant MH111604
NIH R01 Grant DA040621
Michigan State University Honors College Hymen and Miriam Stein Fellowship

Title: Chronic cocaine effects on ER stress in ventral hippocampus

Authors: *A. C. HARLOCK[1], A. L. EAGLE[1], A. ROBISON[2];
[1]Physiol., [2]Michigan State Univ., Michigan State Univ., East Lansing, MI

Abstract: The ventral hippocampus is important in drug behavior, specifically in the drive to seek and take drugs. Over many years, we and others have found that chronic cocaine produces changes in physiology and gene expression in ventral hippocampus neurons, and that these may underlie aberrant cocaine-seeking and reward. We recently found that chronic cocaine increases calreticulin mRNA and protein in ventral hippocampus neurons. Calreticulin is an endoplasmic reticulum (ER) chaperone protein that is important in binding to misfolded proteins and preventing them from being exported from the ER. Misfolded proteins can cause the unfolded protein response (UPR), a compensatory process initiated by ER stress. However, it is unknown whether cocaine-induced calreticulin is linked to ER stress or whether this alters ventral hippocampus function or drug responses. Thus, we hypothesize that cocaine induces calreticulin in ventral hippocampus neurons to alter the UPR and regulate ER stress. Initially, we examined whether chronic cocaine increases UPR proteins, such as ATF6, phosphorylated IRE1, and PERK, in ventral hippocampus tissue from cocaine-treated mice (compared to saline-treated controls) using Western blotting and immunohistochemistry. Male and female C57Bl6/J mice received 10 days of intraperitoneal injections of 20 mg/kg of cocaine (or saline in controls). 24 hours later, ventral hippocampus tissue was taken for Western blotting for ATF6, PERK, and phosphorylated IRE1. Preliminary experiments suggest that cocaine may be increasing the expression of ATF6, PERK, and phosphorylated IRE1 in the ventral hippocampus. Ongoing experiments are also examining whether calreticulin mediates cocaine effects on UPR proteins, using conditional calreticulin knockout mice (Calrfl/fl:Rosa26eGFP-L10a) and viral-mediated overexpression of calreticulin (AAV-CMV-mCalr) in ventral hippocampus neurons to identify whether calreticulin is necessary and sufficient for cocaine-induced increases in UPR proteins. These preliminary findings suggest that chronic cocaine may be producing ER stress, which may underly altered excitability and dysfunction in hippocampal neurons that could lead to aberrant drug seeking.

Disclosures: A.C. Harlock: None. **A.L. Eagle:** None. **A. Robison:** None.

Poster

PSTR167. Cocaine: Cell Signaling, Circuitry, and Neurophysiology

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR167.14/NN5

Topic: G.09. Drugs of Abuse and Addiction

Support: NIH Grant MH111604
NIH Grant DA040621

Title: Cocaine regulates hippocampal physiology and behavior via calreticulin

Authors: *A. L. EAGLE[1], H. M. KUHN[1], B. R. MURRAY[1], M. DYKSTRA[3], R. M. BASTLE[4], M. A. DOYLE[5], J. HE[2], I. MAZE[4], A. J. ROBISON[1];
[1]Physiol., [2]Biochem., Michigan State Univ., East Lansing, MI; [3]Univ. of Michigan, Ann Arbor, MI; [4]Icahn Sch. of Med. At Mount Sinai, Ossining, NY; [5]Vanderbilt Univ., Nashville, TN

Abstract: Ventral hippocampus neurons that send axonal projections to NAc (vHPC-NAc) are critical to cocaine seeking and reward. However, how drugs like cocaine modulate the physiology and gene expression within this neural circuit is unknown. We have discovered that chronic cocaine decreases the excitability of vHPC-NAc neurons *ex vivo* in male mice. Furthermore, cocaine produces this change in a ΔFosB-dependent manner. In our prior studies and current experiments, we show that ΔFosB expression in vHPC-NAc mediates cocaine seeking and reward in male mice. Similarly, cocaine regulates gene expression via ΔFosB in these neurons. Using innovative retrogradely expressing HSVs to drive the Cre-dependent expression of GFP-tagged ribosomes in vHPC-NAc neurons of *Rosa26*$^{eGFP/L10a}$ male mice along with circuit-specific TRAPSeq (Translating Ribosomal Affinity Purification followed by 3rd-genreration sequencing) we identified an endoplasmic reticulum (ER) resident chaperone protein, calreticulin, as a ΔFosB-dependent transcriptional target for cocaine in vHPC-NAc neurons. This finding was further validated by qPCR, Western blotting, immunohistochemistry, and ChIP. We next used a conditional calreticulin knockout mouse line (*Calr*$^{fl/fl}$*: Rosa26*$^{eGFP/L10a}$) and INTERSECT viral strategy, as well as calreticulin overexpression AAVs, to identify whether calreticulin was necessary and sufficient for cocaine behaviors and cocaine-induced decreases in vHPC-NAc excitability. We found that ΔFosB-dependent calreticulin in vHPC was indeed regulating cocaine reward and cocaine-induced changes in vHPC excitability. These studies have elucidated a novel transcription-to-ER pathway by which cocaine regulates the physiology of hippocampal neurons that underlie cocaine seeking and reward.

Disclosures: A.L. Eagle: None. **H.M. Kuhn:** None. **B.R. Murray:** None. **M. Dykstra:** None. **R.M. Bastle:** None. **M.A. Doyle:** None. **J. He:** None. **I. Maze:** None. **A.J. Robison:** None.

Poster

PSTR168. Neural Mechanisms for Choice

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR168.01/NN6

Topic: H.03. Decision Making

Support: Center on Compulsive Behaviors Fellowship

Title: Testing the role of the subthalamic nucleus in memory-guided decisions

Authors: *K. SUNDBY[1], V. SREEKUMAR[2], K. A. ZAGHLOUL[3];
[1]Natl. Inst. of Neurolog. Disorders and Stroke, Bethesda, MD; [2]The Intl. Inst. of Information Technol. Hyderabad, Telangana, India; [3]Surgical Neurol. Br., Natl. Inst. of Neurolog. Disorders and Stroke, NIH, Bethesda, MD

Abstract: Effective decision-making often varies across contexts. As a result, we rely on context memory and past experiences to guide decisions. The subthalamic nucleus (STN) is thought to contribute to decision-making by modifying decision thresholds and withholding actions during uncertainty. Although the STN is often implicated in action-based decisions, less is known about its role in memory-guided decisions. Akin to selecting actions, we hypothesize that the STN is involved in non-motor decisions that are context dependent and require memory. To test this, we recorded intraoperative STN electrophysiology in patients receiving deep brain stimulation for Parkinson's disease as they performed a novel context memory task. The task required patients to learn the appropriate decision between arbitrary categories (cutlery vs. animals and houses vs. faces). These decisions were experienced in two different contexts (beach vs. forest). Importantly, for one decision pair, the decision was context dependent, meaning the correct response differed between the two contexts. We found that patients (n=23) were slower and less accurate when performing a context-dependent decision. Additionally, preliminary analyses reveal heightened STN beta power and an increased spike rate for context dependent decisions, neural responses often associated with action-withholding and conflict, respectively. The current results extend prior work implicating the STN in non-motor decision-making and raise important questions as to how memory systems interact with the basal ganglia to guide decisions.

Disclosures: K. Sundby: None. **V. Sreekumar:** None. **K.A. Zaghloul:** None.

Poster

PSTR168. Neural Mechanisms for Choice

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR168.02/NN7

Topic: H.03. Decision Making

Support: NIH R01 EY-022411

Title: Microstimulation in monkey subthalamic nucleus suggests multiple roles in forming perceptual decisions

Authors: *K. ROGERS, J. I. GOLD, L. DING;
Neurosci., Univ. of Pennsylvania, Philadelphia, PA

Abstract: The subthalamic nucleus (STN) is a part of the indirect and hyperdirect pathways of the basal ganglia (BG) and is believed to be involved in perceptual decision-making, but its exact role is currently unknown. Three main hypotheses have been proposed to explain STN's role in perceptual decision-making, via its effects on BG output activity in the substantia nigra pars reticulata (SNr): 1) STN neurons aggregate evidence for all alternatives to provide a normalization signal (Bogacz and Gurney, 2007); 2) STN provides transient suppression, preventing impulsivity, that disappears over time for both choices (Ratcliff and Frank, 2012); and 3) STN provides sustained suppression until enough evidence has been accumulated to overcome this inhibition and trigger a decision commitment (Wei, et al. 2015).
To test for causal relationships between STN activity and decision behavior, we utilized electrical microstimulation at STN sites in two monkeys performing a visual motion direction-discrimination saccade task. The monkeys made saccades at a self-chosen time to indicate their perception of the global motion direction of a random-dot kinematogram. For each trial, the motion strength and direction were chosen randomly from 5 values and two directions, respectively. The monkey was given a liquid reward for a correct choice. Microstimulation was delivered as a train of biphasic electrical pulses from motion onset to saccade onset (up to 50 μA, 200 Hz). Trials with and without microstimulation were randomly interleaved.
In the 57 microstimulation sessions (n=28 for monkey Cy and 29 for monkey Fa), we observed several effects on choice and reaction time (RT), including: 1) a change in slope of the (logistic) psychometric function in 27 sessions, primary as a reduction (n=22); 2) a horizontal shift in the psychometric function, indicating a choice bias, in 24 sessions, primarily favoring the contralateral choice (n=17); 3) changes in both slope and choice bias (n=15); and 4) changes in RT for at least one choice (n=45). We also fitted a drift-diffusion model to the choice and RT data and observed that multiple model components are needed to explain the microstimulation effects. These results suggest that STN is involved in complex computations for perceptual decision-making. They also highlight the need to create new and/or revised models of the BG to account for the different roles of STN in perceptual decision-making.

Disclosures: K. Rogers: None. **J.I. Gold:** None. **L. Ding:** None.

Poster

PSTR168. Neural Mechanisms for Choice

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR168.03/NN8

Topic: H.03. Decision Making

Title: Differences in sense of agency evaluated by intention binding effect under flat reward condition.

Authors: *S. NOZAWA[1], K. MOGI[2,1];
[1]Dept. of Gen. Systems Studies, The Univ. of Tokyo, graduate school of Arts and Sci., Tokyo, Japan; [2]Sony Comp Sci. Lab., Shinagawa-Ku, Japan

Abstract: Intuitively, will initiates action, a fact that seem almost trivial in the folk psychology of daily life (Nichols 2004). The detailed mechanism of how conscious will effects bodily movements is elusive, as one aspect of the mind-brain problem (Sperry 1952). Correlated brain activities precede the conscious perception of voluntary action as shown in the readiness potential, suggesting that free will is an illusion in terms of causation (Libet 1985), although the interpretations of data are still controversial (Trevena and Miller 2010). It is interesting to study the 'sense of agency' (SoA), the feeling that accompanies one's voluntary action (Moore 2016). In previous studies, the SoA is typically measured implicitly and quantitatively. When a particular action, e.g. a button press, produces outcomes such as an auditory tone, the perceived time of the action shifts towards the time of a subsequent outcome, a paradigm . called intentional binding or action binding. Here we investigate the nature of SoA using a simple gambling task comprising two conditions, namely the 'one box'(O) and 'four boxes'(F) conditions. In the O condition, the picture of one box is presented to the subject. The subject can choose to bet or to escape. When the subject made the bet, 1 unit of resources was taken away. The probability of getting reward was set to be 0.25. Winning the bet was rewarded with 4 units. In the F condition, the picture of four boxes was presented. The subject could choose to bet or not in the same way as in the O condition, by selecting one box to be opened, or escaping. Only one box out of four boxes was rewarded with 4 units. In both O and F conditions, the expectation value is the same (zero). In this flat reward paradigm (Onzo and Mogi 2005), the betting rate in the F condition was significantly higher than in the O condition. In the context of the intentional binding paradigm, we will discuss the function of SoA and the difference of behavior under the statistically equivalent but differently embodied conditions of O and F. Based on the results, we would discuss the neural correlates of SoA, including areas such as the temporoparietal junction and angular gyrus (Bayer et al. 2018, Hughes 2018, Zito et al. 2020).

Disclosures: S. Nozawa: None. **K. Mogi:** None.

Poster

PSTR168. Neural Mechanisms for Choice

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR168.04/NN9

Topic: H.03. Decision Making

Support: NIH Grant RF1-AG067011

Title: Characterizing age-related differences in neural responses during the ultimatum game

Authors: *D. SAZHIN[1], J. WYNGAARDEN[2], O. ZAFF[2], R. LUDWIG[2], M. COLLINS[2], C. SHARP[2], A. DACHS[2], A. NAMBIAR[2], J. DENNISON[3], M. DRAYTON[2], T. TROPEA[2], J. A. CLITHERO[5], T. GIOVANNETTI[2], D. V. SMITH[4];
[1]Temple Univ., Aldan, PA; [2]Temple Univ., Philadelphia, PA; [3]Temple Univ., Philadelphia, NJ; [4]Dept. of Psychology & Neurosci., Temple Univ., Philadelphia, PA; [5]Econ., Univ. of Oregon, Eugene, OR

Abstract: When people receive unfair offers, they tend to reject others even at their own expense (Güth et al., 1982), eliciting activation in the anterior insula (Sanfrey, 2003) and the ventral striatal (Strobel et al., 2011). Nonetheless, it remains poorly understood how aging is associated with changes in economic decision making and, in this investigation, we study the relations between aging and bargaining choices. Participants (N = 20) underwent fMRI while performing the Ultimatum (Güth et al., 1982) as a recipient. The participant accepted or rejected offers (m$16-32) from a random past participant (social condition), or a computer (nonsocial condition). If accepted, both players receive the proposed division; if rejected, neither player gets money. Offers were 5, 10, 25, or 50 percent of the endowment ($16-32). fMRI data was analyzed via a general linear model approach in FSL. Whole brain results were thresholded with a cluster-defining threshold of Z > 3.1, correcting across the whole brain (FWE < 0.05). Using a logistic regression model, our preliminary analyses indicated that participants rejected unfair offers more frequently (B = 0.60, SE = 0.05, t= 11.36, p<.001). Next, our preliminary whole-brain results suggest that social versus nonsocial decisions are associated with enhanced right temporoparietal junction activation. Future analyses will investigate brain activation and connectivity patterns associated with social versus nonsocial decisions, age differences, and risk for financial exploitation. Overall, this study may help identify factors that increase the risk for financial exploitation in older adults.

Disclosures: D. Sazhin: None. **J. Wyngaarden:** None. **O. Zaff:** None. **R. Ludwig:** None. **M. Collins:** None. **C. Sharp:** None. **A. Dachs:** None. **A. Nambiar:** None. **J. Dennison:** None. **M. Drayton:** None. **T. Tropea:** None. **J.A. Clithero:** None. **T. Giovannetti:** None. **D.V. Smith:** None.

Poster

PSTR168. Neural Mechanisms for Choice

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR168.05/NN10

Topic: H.03. Decision Making

Title: The influence of social isolation on patch-leaving foraging decisions in rats

Authors: *Y. DAI, J. R. HINMAN;
UIUC, Champaign, IL

Abstract: Rats are a highly social species, often living in colonies of up to hundreds of individuals. Yet laboratory rats are regularly housed individually despite a large body of literature demonstrating the detrimental effects of such housing. Social isolation has been associated with altered stress responses and impaired cognitive processes, including impacts on decision-making, learning, and memory. Rats raised in social isolation tend to display impulsivity in delay discounting, opting for smaller, immediate rewards. We investigated the impact of post-weaning social isolation on a recently described patch-foraging task for rats (Garcia et al., 2023 bioRxiv). Male (n = 18) and female rats (n = 18) were either singly housed

(SH) or group housed (GH) in same-sex groups of three starting at p24 for four weeks prior to the initiation of behavioral testing. The task arena consists of two open-fields (75cm x 75cm) as patches with depleting rewards rates and a connecting corridor that allows for the manipulation of the travel time between patches. After entering a patch, animals are free to leave the patch at any time in order to travel to the other patch which will begin with its reward rate reset upon entry. The task mimics aspects of natural patch foraging behaviors, in which animals generally follow the marginal value theorem (MVT), although most animals in both the wild and laboratory tend to overstay in patches beyond the optimal leaving time. Both SH and GH rats learned to leave the patches close to the optimal time, but SH and GH of both sexes overstay beyond the optimal leaving time. Surprisingly, GH animals overstayed significantly longer compared to SH animals, which results in SH animals acquiring slightly greater amounts of reward compared to GH animals. As GH animals typically perform better than SH animals on cognitive tasks, the greater departure from optimal performance observed in the GH rats begs many questions. To begin identifying the neural underpinnings of this group difference we euthanized the rats following a final testing session in the patch leaving task and processed their brains for the immediate early gene c-fos to identify regional differences in neural activation. Prior studies have implicated the anterior cingulate cortex in patch leaving decision making based on MVT. One interpretation of the shorter overstaying duration exhibited by the SH animals is that they exhibit greater impulsivity. This can be observed in delay discounting paradigms, which generally involves the orbitofrontal cortex. Our study counterintuitively suggests that increased impulsivity of SH rats may yield closer to optimal performance in a patch leaving foraging task.

Disclosures: Y. Dai: None. **J.R. Hinman:** None.

Poster

PSTR168. Neural Mechanisms for Choice

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR168.06/NN12

Topic: H.03. Decision Making

Support: R01 MH112688

Title: Probing the role of prefrontal cortex, hippocampus, and dorsolateral striatum on decision-making in complex spatial environments

Authors: *U. MUGAN[1], S. L. HOFFMAN[2], A. D. REDISH[1];
[1]Neurosci., Univ. of Minnesota, Minneapolis, MN; [2]Intramural Res. Program, NIDA, Baltimore, MD

Abstract: Studies on decision-making suggest that adaptive decisions arise from multiple systems in the brain optimized for different situations and contexts including a flexible system that uses action-outcome expectancies, and an inflexible system that uses situation-action

associations. Computational modeling has suggested that the complexity of an environment changes the usefulness of these strategies. This raises two questions: 1) how does different neural circuitry represent structurally different environments, and 2) how do interactions between brain regions change to promote adaptive behaviors in differently-complex environments.
Three regions have been proposed to make distinct contributions to decision-making: hippocampus (HC) - encoding the state-space and enabling flexible plans, dorsolateral striatum (DLS) - encoding less flexible action chains, and prefrontal cortex (mPFC) - encoding the task-space and strategies. We examined neural activity in rats on a left/right/alternation foraging task by simultaneously recording across these structures with silicon probes and disrupting mPFC with DREADDs (CAMKIIa-h4MDi) in a separate cohort of rats. Rats ran through a central path (parametrically changed to modulate environmental structure) and either turned left or right for a food reward. Reward contingency changed twice through each 45 min daily session. Each task structure was repeated once in a pair of days.
Chemogenetic disruption of mPFC decreased vicarious trial and error (VTE) events and exploration, which rebounded the next day when rats experienced a repeat of the task structure with no disruption. Moreover, mPFC disruption caused behaviors to adapt more slowly to reward changes. HC, mPFC, and DLS task-relevant neural activity patterns varied by environment complexity. In HC, more complex environments increased the spatial information of neural activity, resulted in more non-local activity, and led to longer sharp wave ripple (SWR) complexes. In DLS, classical task-bracketing - preferential firing at the start and end of a lap - only appeared in simple environments. In mPFC, neural changes associated with strategy switches preceded behavioral adaptations. Both mPFC and DLS represented simple and complex environments differently, suggesting that presence of intermediary decision points impacted subgoal representations. Together, these data suggest environmental structure impacts task-relevant representations, and mPFC engagement, which is hypothesized to be important for strategy setting and initiation of deliberative sequences, is particularly crucial in complex environments.

Disclosures: U. Mugan: None. **S.L. Hoffman:** None. **A.D. Redish:** None.

Poster

PSTR168. Neural Mechanisms for Choice

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR168.07/NN13

Topic: H.03. Decision Making

Support: NSF CCF 2211645

Title: Joint identification of individual and group level functional connectomes during Flanker task

Authors: *S. AVIYENTE[1], A. KARAASLANLI[2];
[1]Michigan State Univ., East Lansing, MI; [2]Univ. of Michigan, Ann Arbor, MI

Abstract: Identifying principles of brain function linking disruptions in normative functional connectome (FC) behavior to neurological and psychiatric disorders is an emerging and challenging neuroscience research problem. Most of the current work has focused on providing a cumulative understanding of neural mechanisms underlying higher-order cognitive functioning. Recently, there has been a research paradigm shift from group-level inference to individual-level prediction. In particular, it is important to quantify individual differences for both health and disease. In this study, we focus on inferring the functional connectomes jointly at the individual and group levels by a multiview graph learning (mvGL) method. The proposed method extends the current literature on learning smooth graph structures from observed data to multiple graphs, where both individual functional connectomes and a consensus connectome are learned. This framework is applied to electroencephalogram (EEG) data collected from twenty subjects performing a cognitive control-related error processing task. During recordings, subjects perform a letter version of the speeded reaction Flanker task, where a string of five letters, either congruent (e.g., SSSSS) or incongruent (e.g., SSTSS), is shown at each trial. Subjects use a standard mouse to react to the center letter and the goal is to capture the Error-Related Negativity (ERN) after an error response. In this study, data from error trials are employed for graph learning with the goal of inferring the FC in the theta band for each subject. Since subjects perform the same cognitive task, it is assumed that there is a common graph structure across subjects. By applying mvGL, the learned FC graphs are ensured to be similar to each other and the learned consensus graph represents the common structure across subjects. The topology of the learned connectomes is further characterized through modularity-based community detection. The results indicate that there's a consensus community centered around frontal-central regions consistent with prior work indicating the increased activation of medial prefrontal cortex (mPFC) during cognitive control. In addition, there are group level communities centered around the left and right lateral prefrontal cortices and the visual and motor regions.

Disclosures: **S. Aviyente:** None. **A. Karaaslanli:** None.

Poster

PSTR168. Neural Mechanisms for Choice

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR168.08/NN14

Topic: H.03. Decision Making

Support: Wellcome 214352/Z/18/Z
Wellcome 219627/Z/19/Z
Gatsby Charitable Foundation GAT3755
European Research Council Consolidator #864912

Title: An assay for investigating goal switching during hunting and escape behaviour

Authors: ***S. F. OLESEN**[1], N. J. MILLER[1], B. CRUZ[3], G. LOPES[3], N. BURGESS[2], T. BRANCO[1];

[1]Sainsbury Wellcome Ctr., [2]UCL Inst. of Cognitive Neurosci., Univ. Col. London, London, United Kingdom; [3]NeuroGEARS Ltd, London, United Kingdom

Abstract: Consuming food and evading threat are fundamental behaviours that compete, and switching between them is essential for survival. An animal engaged in hunting must quickly stop and switch to a defensive strategy if it faces imminent threat. The neural mechanisms for fast interruption and switching between natural goal-directed behaviours have not been identified. In mice, hunting relies on vision-guided prey pursuit, while escape from threat uses spatial memory to reach shelter. During both behaviours, however, mice accurately orient to the current goal: when hunting crickets mice maintain orientation towards the moving prey; when escaping from threat, they orient to the shelter location. Therefore, to switch from hunting to escape, mice must break vision-guided orienting actions and initiate a new memory-guided orientation movement. The different orienting actions are likely carried by the same effector circuits, such as the superior colliculus, suggesting that these circuits must switch between controllers. We have developed behavioural paradigms in freely behaving and head-fixed mice to investigate the neural mechanisms behind goal-orienting during hunt-to-escape switching. First, we implemented a paradigm where freely behaving mice hunt live crickets in an arena with a shelter. Presentation of threat stimuli during pursuit bouts elicited escapes to shelter. Hunting mice maintain accurate head orientation towards the moving cricket but break their prey-orientation lock in response to threat stimuli, orienting instead towards the shelter. This orientation break is fast (~350ms) and the accuracy to the goal is maintained, indicating a rapid and accurate switch between orienting goals. We next designed a platform for implementing an equivalent behavioural paradigm in head-fixed configuration. We built a virtual reality setup where mice are head-fixed onto a bearing that allows 360-degree yaw rotation, along with a motorized arm holding a live cricket. The virtual environment has a shelter, and the cricket movements are controlled in closed-loop, responding to the movement of the mouse and reacting according to pre-programmed rules. We show that head-fixed mice orient towards, chase, and consume the tethered cricket, and escape to the virtual shelter when presented with threatening stimuli, like natural hunting and escape behaviours. We aim to combine these two behavioural paradigms to study hunting-to-escape orientation switching under natural and controlled conditions, and use complementary recording and neural activity manipulation techniques to determine the underlying neural mechanisms.

Disclosures: S.F. Olesen: None. **N.J. Miller:** None. **B. Cruz:** None. **G. Lopes:** None. **N. Burgess:** None. **T. Branco:** None.

Poster

PSTR168. Neural Mechanisms for Choice

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR168.09/NN15

Topic: H.03. Decision Making

Support: NIH Grant R01MH122513

Title: Post-choice dynamics in fronto-insular network during perceptual decision making

Authors: *S. GHERMAN[1], N. MARKOWITZ[2], G. TOSTAEVA[3], E. ESPINAL[4], A. MEHTA[3], S. KELLY[5], R. O'CONNELL[6], S. BICKEL[7];
[1]The Feinstein Inst. for Med. Res., Manhasset, NY; [2]North Shore Hosp., Greenwich, CT; [3]Feinstein Inst. for Med. Res., Manhasset, NY; [4]Feinstein Inst. for Med. Res., Philadelphia, PA; [5]Sch. of Electrical and Electronic Engin., Univ. Col. Dublin, Dublin, Ireland; [6]Trinity Col. Dublin, Dublin, Ireland; [7]Neurosurg. - Neurol., Feinstein Inst., Manhasset, NY

Abstract: Human neuroimaging and intracranial EEG (iEEG) work has previously implicated fronto-insular regions in performance monitoring processes (i.e., selective responses to self-detected errors and/or negative feedback). Concurrently, some of the same regions have been implicated in accumulating abstract sensory evidence during perceptual decision making. In a previous study from our lab that recorded iEEG from human subjects, we identified a widely distributed network where activity during evidence presentation exhibited key characteristics associated with abstract evidence accumulation. Here, we investigate post-choice dynamics at these locations to disentangle potentially distinct contributions of the identified regions to the decision making process. Presurgical epilepsy patients (N=23) judged the direction (up vs. down) of random-dot stimuli and reported their choice with a speeded button press. Feedback on the accuracy of the responses was provided after each trial. We analyzed high frequency activity (70-170 Hz) time-locked to the presentation of sensory evidence and choice commitment (i.e., motor response). During evidence presentation, sites categorized as potential abstract evidence accumulators exhibited gradual ramping responses that peaked just before commitment to choice, scaled positively with the strength of sensory evidence, increased at a rate that predicted reaction time, and were independent of motor effector. Intriguingly, in the time interval immediately following subjects' commitment to choice (and before feedback was provided), a subset of these sites exhibited activity that persisted after the response on error but not correct choices. These were located predominantly in regions of the frontal cortex and the dorsal anterior insula, including regions that have been previously implicated in performance monitoring processes. We further investigate the possibility that the observed pattern reflects processes related to error-detection vs. post-choice evidence accumulation. These preliminary findings provide insights into the complex functional and neurophysiological dynamics involved in perceptual decision making and post-choice processing.

Disclosures: S. Gherman: None. **N. Markowitz:** None. **G. Tostaeva:** None. **E. Espinal:** None. **A. Mehta:** None. **S. Kelly:** None. **R. O'Connell:** None. **S. Bickel:** None.

Poster

PSTR168. Neural Mechanisms for Choice

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR168.10/NN16

Topic: H.03. Decision Making

Support: ASAP Grant 020505

Title: Locus Coeruleus activity facilitates a switch between strategy networks in a model of cognitive flexibility

Authors: *R. E. S. PARKER[1,2,3], M. P. CONTRERAS[1,2], J. BRAUN[3], M. PRIGGE[1,2,4];
[1]Res. Group Neuromodulatory Networks, Leibniz Inst. for Neurobio., Magdeburg, Germany;
[2]Aligning Sci. Across Parkinson's (ASAP) Collaborative Res. Network, Chevy Chase, MD;
[3]Fakultät für Naturwissenschaften (FNW), Otto Von Guericke Univ., Magdeburg, Germany;
[4]Ctr. for Behavioral Brain Sci., Magdeburg, Germany

Abstract: One sentence Summary: Noradrenergic Nucleus exhibits learned activity dynamics that coincide with choosing a rewarding strategy. The Locus Coeruleus (LC), the only source of the neuromodulator noradrenaline in the brain, is thought to play a key role in cognitive flexibility by resetting previously learned reward associations, so that new ones may be learned. The LC is also one of the earliest nuclei affected in Parkinson's Disease (PD), therefore understanding the contribution of the LC in this cognitive capability is of major relevance. Here we aim to map changes in LC activity during strategy-switching behavior in mice. To achieve this aim, we used fiber photometry to record calcium responses in the LC of freely moving mice performing a cognitive flexibility task. In the task, mice switched between two rules to earn a water reward. The first rule required turning in a specific direction while the second rule required navigating to a specific location. Animals received daily sessions to learn the rule until reaching a performance criterion (80% correct). This rule was then reversed and reversed again after reaching criterion with the new rule. On each trial, animals were confined to the start arm until the door opened and released them to freely choose between two opposing reward arms where animals poked to receive water. Trials were recorded, labeled (DeepLabCut), and passed through a semi-supervised network (A-SOID) to extract behavioral poses. First, we found an increase in LC activity time-locked to the nose-poke that developed over the course of learning and vanished after reversal. Furthermore, we found that the highest peaks of LC activity, other than reward, at first occurred scattered through the maze. After learning, the highest peaks in each trial were concentrated at the door of the start arm. After reversal, the peaks of activity were redistributed near the center of the maze, where animals presumably gather spatial information to choose the correct arm. To explore changes in behavior after reversal, we distinguished three behavior categories: moving, searching, and drinking. Results showed that animals increased the time spent searching after reversal and that there was an increase in LC activity during transitions from searching to moving. This behavioral-transition increase was only observed after the reversal session and did not diminish with further training, agreeing with the notion that the LC contribution to optimal cognitive flexibility occurs in concert with changes in behavioral states while also suggesting that the LC facilitates the switch between multiple discrete strategies, rather than the forgetting of previously learned strategies.

Disclosures: R.E.S. Parker: None. **M.P. Contreras:** None. **J. Braun:** None. **M. Prigge:** None.

Poster

PSTR168. Neural Mechanisms for Choice

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR168.11/NN17

Topic: H.03. Decision Making

Support: NIH Grant R15DA046797

Title: The role of lateral orbitofrontal cortex to ventral pallidum circuitry in sensitivity to delayed and immediate punishment during decision-making

Authors: *G. L. MINNES, E. A. DUECKER, S. C. LOWE, V. E. WILLIAMS, A. G. CRICHTON, N. W. SIMON;
Univ. of Memphis, Memphis, TN

Abstract: Cost/benefit decision-making involves the assessment of all potential positive and negative outcomes to maximize reward and minimize punishment. However, punishments that occur later in time are often underestimated, which can lead to maladaptive decision-making. Sensitivity to delayed punishment can be measured using the Delayed Punishment Decision-making Task (DPDT), which reveals that rats avoid rewards accompanied by immediate punishment, then shift preference toward that option when punishment is delayed. Previous work has shown that sensitivity to delayed punishment is regulated by the lateral orbitofrontal cortex (LOFC), but little is known about the specific LOFC circuits that regulate evaluation of delayed punishment. One projection of interest is LOFC to ventral pallidum (VP), a region in the basal ganglia involved in regulating reward and punishment sensitivity, risk-taking, and drug-seeking behaviors. Here, we investigated the role of LOFC-VP circuitry in sensitivity to punishment occurring after different time intervals in male and female rats. First, we determined the effects of VP inactivation with baclofen/muscimol on DPDT, then used inhibitory Designer Receptors Exclusively Activated by Designer Drugs (DREADDs) to suppress activity in LOFC-VP circuit during DPDT. VP inactivation (n=5) produced a trend toward reduced choice of punished rewards regardless of punishment delay. Chemogenetic inactivation of the LOFC-VP circuit (n=5) led to inability to adapt to changes in delay, possibly a result of impaired behavioral flexibility. Understanding the role of specific LOFC circuits in sensitivity to punishment may lead to novel circuit-level treatments to improve aberrant decision-making in substance use disorders and other disorders.

Disclosures: G.L. Minnes: None. **E.A. Duecker:** None. **S.C. Lowe:** None. **V.E. Williams:** None. **A.G. Crichton:** None. **N.W. Simon:** None.

Poster

PSTR168. Neural Mechanisms for Choice

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR168.12/NN18

Topic: H.03. Decision Making

Support: NIH Grant S10OD023675-01 (PI: AC Mar)

Title: Prefrontal dopamine and serotonin in a novel Delay and Reward Discounting task for rodents

Authors: *B. GAMALLO LANA[1], F. ARTIGAS[2], A. C. MAR[3];
[1]NYU Langone Hlth. - Neurosci. Inst., New York, NY; [2]IIBB-CSIC ESQ2818002D, Barcelona, Spain; [3]New York Univ. Sch. of Med., New York, NY

Abstract: Delay discounting (DD) is a measure of preference for rewards delivered immediately or sooner than more delayed rewards. Discounting is important in daily decision making and higher levels of DD have been linked to a number of psychological disorders, such as ADHD and substance abuse disorders. DD is associated with a number of neurochemical systems, including serotonin (5-HT) and dopamine (DA). Reduced 5-HT function has been shown to decrease discounting rates, while higher 5-HT levels increase the 'willingness' to wait. Conversely, low DA is often related to increased discounting. The prefrontal cortex (PFC) contributes to value-based decision-making by representing and comparing the subjective value of reward options and guiding flexible choices. The interaction and modulation of DA and 5-HT within the PFC during DD performance is not yet understood. DD in rodents is commonly assessed by two-choice operant tasks in which animals select between a small, immediate reward, and a larger, delayed reward. In most protocols, the magnitude and location of the small, immediate reward option typically remains fixed throughout a given session or experiment. This configuration makes it difficult to separate either sensitivity to differences in reward amount or side bias from actual choice preference based on value. In this study, we have developed and validated a novel Delay and Reward Discounting (DRD) task for rodents. On each trial, two visual stimuli are presented side-by-side on a touch screen. Animals are then freely allowed to touch one and, after a programmed delay period, a liquid reward is delivered. Reward size and delay on a given trial are uniquely dependent on the chosen stimulus. Each combination of reward size (15, 25, 35, 50µL) and delay (0, 2, 4 or 6s) are represented by the visual properties of 16 different images. Side biases can be corrected by titrating forced choice trials. Multilevel logistic regression modeling was applied to the choice data of 25 mice to estimate the separate parametric contributions of sensitivity to reward magnitude ratio and relative delay. We observed that although mice show an overall higher discounting based on delay length, mice present high heterogeneity and individual differences in reward and delay discounting rates. We expressed genetically encoded GRAB sensors in medial PFC and observed a 5-HT reward-dependent signal after collection and a delay-dependent signal in the delay to reward period. Our results suggest that DRD may be a powerful translational tool for elucidating the mechanisms of DD, impulsivity and decision-making processes involving executive function.

Disclosures: B. Gamallo Lana: None. **F. Artigas:** None. **A.C. Mar:** None.

Poster

PSTR168. Neural Mechanisms for Choice

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR168.13/NN19

Topic: H.03. Decision Making

Support: NIMH Grant R01MH119086

Title: The influence of cognitive fatigue on physical effort-based decision-making

Authors: *M. H. DRYZER;
Biomed. Engin., Johns Hopkins Univ., Baltimore, MD

Abstract: Fatigue is a lack of energy or motivation that can be caused by physically or cognitively demanding tasks. It is often anecdotally reported that fatigue in one effort domain (e.g., cognition) can impact judgments of effort in another domain (e.g., physical). Despite the supposed cross-talk between fatigue domains, little is known about how fatigue in one domain influences decisions to exert in another domain. In this experiment, we characterized individuals' subjective preferences for physical effort and examined how these preferences, and associated brain activity, changed after they were cognitively fatigued. To achieve this, we scanned participants with functional magnetic resonance imaging (fMRI) while they made decisions about exertion of physical effort (i.e., grip exertion), before and after they performed bouts of cognitively fatiguing exertion (i.e., working memory). Participants first learned to associate their physical grip-exertion with a numerical scale ranging from zero to one hundred, relative to their maximum physical exertion. They then performed a series of risky binary gambles involving varying levels of physical exertion. During each choice, participants decided to risk equal probability of high physical exertion or rest, or forgo any risk and select a guaranteed lower level of exertion. After completing a baseline session of choices for physical effort, participants performed a series of cognitively fatiguing n-back working memory trials. Following these cognitive fatigue bouts, they repeated the same physical exertion choices as in the baseline state. In a baseline state, we found that participants exhibited convex subjective effort value functions, indicating that the marginal subjective values of physical effort increased as more effort was required. When participants were in a cognitively fatigued state, the marginal subjective value of physical effort became more pronounced, as indicated by an increased convexity in the subjective effort value function. Analyses of the neuroimaging data revealed that right insula, while insensitive to effort value in a baseline state, was sensitive to cognitive fatigue-induced changes in physical effort value. Additionally, we found that right dorsolateral prefrontal cortex (dlPFC) encoded increasing cognitive load due to repeated cognitive exertion during the n-back task. A psychophysiological interaction analysis showed that changes in right insula activity between baseline and fatigued states were positively correlated with fatigue-related changes in right dlPFC. These results provide an account of how cognitive fatigue influences decisions to exert physical effort.

Disclosures: M.H. Dryzer: None.

Poster

PSTR168. Neural Mechanisms for Choice

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR168.14/NN20

Topic: H.03. Decision Making

Support: NARSAD Young Investigator Award
NIH F32MH110135
R01DA038063
5R01DA038063

Title: Neural mechanisms underlying the prospective estimation of self-control costs

Authors: *C. RAIO[1], L. LEONE[2], A. KONOVA[3], P. W. GLIMCHER[4];
[1]Psychiatry, NYU Sch. of Med., New York, NY; [2]Psychology, Univ. of Texas at Austin, Austin, TX; [3]Psychiatry and UBHC, Rutgers Univ. at New Brunswick, New Brunswick, NJ; [4]Neurosci., New York Univ., New York, NY

Abstract: Failures of self-control have significant implications for both individual and public health, affecting various economic and health domains. Converging work across cognitive and decision neuroscience has shown that exerting control is registered as cognitively costly. We previously demonstrated that the subjective cost of self-control can be measured behaviorally using a willingness-to-pay mechanism, and further, that stress exposure increases the perceived cost of exercising control. Here, we sought to characterize the neural circuits underlying how these costs are estimated. We tested the hypothesis that prospective control cost estimates will be encoded in anterior prefrontal regions consistent with past precommitment work [e.g. frontopolar cortex (FPC), orbitofrontal cortex (OFC)] as well as regions implicated in encoding the ongoing cognitive cost of control [dorsal anterior cingulate cortex (dACC)], rather than traditional cognitive control regions responsible for deploying control (e.g., dlPFC). Healthy dieters (n=25) first rated snack-foods on health, taste and temptation level in order to select a low, medium and high-tempting food for each individual. Participants then underwent fMRI scanning while completing a self-control choice task. On each trial, participants viewed offers that varied on temptation level (low, medium, high), quantity (small, medium, large) and duration of time they would later need to spend with the food (1-60 min), and for each submitted a willingness-to-pay bid (from a $10 study endowment) to avoid the specified food/time depicted. A realization phase followed the scanning session, during which one trial (bid) was randomly selected and entered into a standard economic auction procedure (BDM), which determined whether time with the tempting food was successfully avoided or not. Brain activity was modeled with a parametric modulator of (raw) bid value during the decision period when participants evaluated how much to pay to avoid control. Higher bids yielded increased activation in FPC, mOFC and dACC, pointing to a central role of these brain regions in prospectively estimating the perceived cost of self-control. There was no measurable change in activation in the dorsolateral prefrontal cortex during bid decisions. Our data suggest that estimating the subjective cost of exercising self-control engage a distinct neural circuit than that traditionally involved in implementing control. Gaining a better understanding of the neural basis of these cost estimates may provide potential neural targets to help improve the success of prospective self-control strategies.

Disclosures: **C. Raio:** None. **L. Leone:** None. **A. Konova:** None. **P.W. Glimcher:** None.

Poster

PSTR168. Neural Mechanisms for Choice

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR168.15/OO1

Topic: H.03. Decision Making

Support: Allen Institute for Neural Dynamics
Howard Hughes Medical Institute

Title: Mesoscale dynamics of neural populations reveal high-dimensional communication across cortical areas

Authors: *L. SUSMAN[1,2], J. ALJADEFF[4], K. SVOBODA[5,6], A. FINKELSTEIN[2,3];
[1]Princeton Univ., Princeton, NJ; [2]Dept. of Physiol. and Pharmacology, Fac. of Med., [3]Sagol Sch. of Neurosci., Tel Aviv Univ., Tel Aviv, Israel; [4]Section of Neurobiology, Div. of Biol. Sci., UCSD, La Jolla, CA; [5]Allen Inst., Seattle, WA; [6]Janelia Res. Campus, Ashburn, VA

Abstract: Many cognitive processes unfold on a wide range of spatial and temporal scales and involve multiple brain areas. Recent studies have begun to characterize spatiotemporal dynamics across the neocortex, however, little is known about the nature of cortex-wide activity at the level of individual cells. To address this gap, we used mesoscale calcium imaging (Sofroniew et al., 2016) to record the activity of up to ~30,000 neurons simultaneously (~1,000,000 neurons total) from over 10 cortical areas, including somatosensory, motor, and high-order visual areas. Cortical activity was recorded while mice performed a goal-directed behavior involving multidirectional tongue-reaching for water rewards. Global cortical activity (across areas) - as well as activity within each cortical area - was high-dimensional, with the variance of activity on each dimension exhibiting a broad spectrum. The spatial scale of activity along the cortical sheet also exhibited a very broad spectrum; specifically, the dimensions with high variance were spatially extended, spanning multiple areas, whereas dimensions with intermediate variance were local. The high-variance dimensions were correlated with behaviorally relevant signals. In contrast, the intermediate-variance dimensions represented the communication subspace (Semedo et al., 2019) across different areas. This communication subspace was also high-dimensional, and its geometry was not aligned with the dominant dimensions of activity within individual brain areas. Our results suggest that cortical activity decomposes into two major components: a subspace of behavior-related activity with broad anatomical distribution, and a high-dimensional subspace representing the flow of information across brain areas.

Disclosures: L. Susman: None. **J. Aljadeff:** None. **K. Svoboda:** None. **A. Finkelstein:** None.

Poster

PSTR168. Neural Mechanisms for Choice

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR168.16/OO2

Topic: H.03. Decision Making

Support: MU-CIN Grant

Title: Role of hippocampus in economic decision making: evidence from intracranial recordings

Authors: *S. GOULIS[1,2], M. VERWOERT[3], M. C. OTTENHOFF[3], L. WAGNER[5], P. KUBBEN[3,6], M. WIBRAL[2,4], C. HERFF[2,3], V. VAN DE VEN[1,2];
[1]Cognitive Neurosci. & Ctr. for Integrative Neurosci., [2]Ctr. for Integrative Neurosci., [3]Dept. of Neurosurgery, Sch. of Mental Hlth. and Neurosciences, [4]Dept. of Microeconomics and Publ. Economics, Sch. of Business and Econ., Maastricht Univ., Maastricht, Netherlands; [5]Academic Ctr. for Epileptology, Kempenhaeghe/MaastrichtUniversity Med. Ctr., Heeze, Netherlands; [6]Academic Ctr. for Epileptology, Kempenhaeghe/MaastrichtUniversity Med. Ctr., Maastricht, Netherlands

Abstract: In certain types of economic decision making, humans exhibit a framing effect in which decisions with identical outcomes are influenced by their presentation either as gains or losses. The amygdala and frontal brain regions have been implicated in this decision bias, mainly through evidence from functional magnetic resonance imaging. Recently, the hippocampus, a brain area traditionally associated with episodic memory, spatial cognition and associative learning, was also found to be involved in economic decision making, especially in the process of value retrieval of specific stimuli. We sought to further investigate the role of hippocampal processing in economic decisions in a framing task via intracranial encephalography recordings in the hippocampi of 8 epileptic patients. Participants were initially endowed a monetary amount (e.g. 100 Euros) and then presented with a safe option to keep part of this amount, which in some trials was framed either as a loss (Lose 60 Euros) or as a gain (Keep 40 Euros), and a gamble option to keep the whole previously endowed amount. Importantly, this task relied on manipulating monetary outcomes and their frame without using episodic stimuli that could drive associative recall. Our results showed that a framing effect was successfully induced across participants, namely that they preferred the gamble option more in the loss frame than gain frame trials, especially when deciding about higher monetary amounts. Hippocampal theta power was substantially modulated during the option deliberation and pre-decision periods relative to pre-trial baseline. Interestingly, this modulation was significantly stronger in the loss frame than gain frame trials in the pre-decision period and particularly in higher monetary amount trials. Additionally, in 5 out of 8 participants we found a hippocampal event-related potential (ERP) time-locked to option presentation onset (~700ms). The ERP peak amplitude was smaller in the loss frame than in the gain frame trials, possibly reflecting reduced gain anticipation for the loss frame trials and in line with a framing effect. Lastly, we investigated the occurrence of hippocampal sharp wave ripples (SWRs) throughout the task and found an increased average SWR rate during decision-relevant periods. Taken together, we demonstrate novel evidence pointing to a hippocampal involvement in a type of economic decision making in the absence of episodic stimuli, and that hippocampal activity during deliberation correlates with a framing effect. These results expand the current view on tasks in which the hippocampus is involved as well as the neural substrates of a framing effect.

Disclosures: **S. Goulis:** None. **M. Verwoert:** None. **M.C. Ottenhoff:** None. **L. Wagner:** None. **P. Kubben:** None. **M. Wibral:** None. **C. Herff:** None. **V. van de Ven:** None.

Poster

PSTR168. Neural Mechanisms for Choice

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR168.17/OO3

Topic: H.03. Decision Making

Support: R01 EY015260
NSFGRFP DGE 1845298

Title: Contributions of sensory adaptation and pupil-linked arousal to perceptual decisions about uncertain and unstable visual stimuli

Authors: *K. D. MCGAUGHEY[1], J. I. GOLD[2];
[1]Univ. of Pennsylvania Neurosci. Grad. Group, PHILADELPHIA, PA; [2]Univ. of Pennsylvania, Univ. of Pennsylvania, Philadelphia, PA

Abstract: Visual decisions often require the accumulation of uncertain sensory evidence over time. To be effective in the real world, where evidence is both uncertain and unstable, the dynamics of this accumulation process must adjust to the level of environmental stability. For example, in increasingly unstable environments where evidence is likely to change at any moment, the time window over which evidence is accumulated should become increasingly narrow. We have shown that humans and monkeys use this kind of flexible evidence accumulation. Here, we investigate the underlying neural mechanisms, focusing on potential contributions from two complementary processes: (1) bottom-up, stimulus-dependent adjustments to the dynamics of adaptation in neurons that encode sensory evidence (e.g., increasingly transient and/or weak responses following increasingly common stimulus changes); and (2) top-down, context-dependent modulations of the temporal dynamics of the evidence-accumulation process by pupil-linked arousal systems (e.g., affecting the time course of accumulation based on learned expectations about the rate of environmental changes).We trained two macaques (n = 35 sessions for Monkey Ch; n = 53 sessions for Monkey An) on a random-dot motion task in which we manipulated context stability via different rates of "change-points," or abrupt switches in motion direction. The monkeys, like humans, tended to accumulate evidence over shorter timescales when change-points occurred more often. We recorded activity of neurons in middle temporal area (MT). Preliminary analyses (*n = 70 single units)* suggest that cortical sensory adaptation was stronger in MT when change-points occurred more often. Differences in sensory adaptation corresponded to differences in behavior, including stronger modulations of neural adaptation on sessions with *vs.* without reliable, stability-dependent adjustments in evidence-accumulation behavior. Additional preliminary analyses suggest that the dynamics of pupil diameter, which are known to encode learned expectations about stimulus dynamics, were also sensitive to the evidence-accumulation process. Specifically, pupil

modulations around behaviorally relevant change-points depended on whether the monkeys tended to use flexible evidence accumulation or not in a given session, suggesting that pupil diameter is sensitive to learned, behaviorally relevant expectations about context stability. Collectively, these results demonstrate that stimulus dynamics can induce adjustments to both bottom-up evidence encoding and top-down arousal modulations that may contribute to flexible perceptual decision-making.

Disclosures: K.D. McGaughey: None. **J.I. Gold:** None.

Poster

PSTR168. Neural Mechanisms for Choice

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR168.18/OO4

Topic: H.03. Decision Making

Support: JSPS 19H05467
JSPS 22H05497
JSPS 22K15226

Title: Neural representations of medial prefrontal cortex neurons projecting to the striatum for diminishing reward-based action switch

Authors: *S. NONOMURA, T. KANEKO, H. AMITA, K. INOUE, M. TAKADA;
Kyoto Univ., Inuyama, Japan

Abstract: Action-outcome contingency, in an unstable and dynamic environment, is gradually changed by repeating an action which makes the outcome value diminish. We often face the situation in which there is a dilemma whether to keep or switch an action that may cause no reward definitively. It is well known that the cortico-basal ganglia circuitry plays a pivotal role in action selection based on the outcome value. However, it remains unknown how neurons giving rise to this circuitry process the information about the action selection with a reward progressively diminishing. Here, we focused on the medial prefrontal cortex (mPFC) neurons projecting to the striatum (Str) and investigated its functional role in diminishing reward-based action switch. To this end, we first developed a novel behavioral task in which rats under a head-fixed condition had to choose a right or left pedal with the corresponding forelimb to acquire a better water reward when the reward value was changed. Reward diminishment occurred when the rats repeatedly chose the same action associated with a certain reward for 5-15 successive trials. The rats successfully switched their actions (i.e., the right to left pedal or the left to right pedal) when the reward diminished. To examine whether the mPFC-Str projection contributes to the action switch after the reward diminishment, we next made ibotenic acid lesions in the bilateral mPFC. After the lesions, the probability of action switch was significantly increased within the first to third trials following the onset of reward diminishment. To define an involvement of the mPFC-Str projection in diminishing reward-based action switch, we finally

identified the activities of mPFC-Str neurons (n = 100) by opt-tagging. Along with the mPFC-Str neurons, activities of striatal indirect pathway neurons (IPNs; n = 34) were analyzed in the same fashion. It was found that both the mPFC-Str neurons and the IPNs showed similar reward outcome responses around the onset of reward diminishment. However, a multiple linear regression analysis with bootstrapping demonstrated that the mPFC-Str neurons mainly encoded negative reward prediction error signal, whereas the IPNs encoded next action switch signal. These results indicate that mPFC-Str neurons and IPNs may cooperatively be involved in signal processing responsible for diminishing reward-based action switch.

Disclosures: **S. Nonomura:** None. **T. Kaneko:** None. **H. Amita:** None. **K. Inoue:** None. **M. Takada:** None.

Poster

PSTR168. Neural Mechanisms for Choice

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR168.19/OO5

Topic: H.03. Decision Making

Support: Wellcome Trust: 217211/Z/19/Z
Wellcome Trust: 224121/Z/21/Z

Title: Parallel frontal cortico-basal ganglia networks balance impulsivity and sensory evidence integration during decision-making

Authors: ***M. LOHSE**, B. J. GONZALES, M. SKRETOWSKA, A. KHILKEVICH, P. WINDMILL, R. A. A. CAMPBELL, T. D. MRSIC-FLOGEL;
Sainsbury Wellcome Centre/UCL, London, United Kingdom

Abstract: Deciding when to act based on ambiguous sensory input requires the balancing of two behavioural strategies: not acting too soon and not missing the moment when sufficient sensory evidence is accumulated. How the brain balances impulsivity and sensory evidence integration to act at the right time is not understood.
We examined this question in mice performing a reaction time task in which they are required to sit still until they detect and report (by licking a reward spout) a sustained change in speed of a drifting grating which fluctuates noisily around a baseline speed of 1Hz. Using measurements and optogenetic manipulations of neural activity, we found that the neurons in the anterior region of secondary motor cortex (MOs) integrate evidence for longer than those in the posterior regions of MOs, and silencing anterior MOs decreases detections of changes in stimulus speed. Conversely, neurons in posterior MOs have reduced firing rates in trials when mice make impulsive licks (i.e., before a sustained change in visual speed), and accordingly silencing posterior MOs increases impulsive lick rates. This reveals functionally specialized circuits separated along the anterior-posterior axis of MOs which causally contribute to either impulsivity control or sensory-to-action transformation that requires evidence integration.

We hypothesized that these subdivisions of MOs may enact the balance between impulsivity and timely action through their topographically organized projections to striatum; the dorsomedial striatum (DMS) receives inputs from the posterior MOs, and the ventromedial striatum (VMS) receives inputs from the anterior MOs. Preliminary results show that silencing D2+ neurons (A2a-cre) in the DMS - but not VMS - increased the impulsive licking, suggesting that the control of impulsivity by posterior MOs is mediated through D2+ neurons in DMS.
We are currently investigating how silencing posterior MOs and anterior MOs affects the representations of impulsivity and evidence integration in DMS and VMS, and early results suggest that silencing of MOs disrupts the integration of sensory evidence in striatum.
Together, these findings suggests that parallel and cell type-specific cortico-striatal circuits distinctly control impulsivity and sensory-to-action transformations, and more generally demonstrate that distinct behavioral strategies can be implemented through parallel cortico-basal ganglia circuits.

Disclosures: M. Lohse: None. **B.J. Gonzales:** None. **M. Skretowska:** None. **A. Khilkevich:** None. **P. Windmill:** None. **R.A.A. Campbell:** None. **T.D. Mrsic-Flogel:** None.

Poster

PSTR168. Neural Mechanisms for Choice

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR168.20/OO6

Topic: H.03. Decision Making

Support: T32 DA037183
R01 MH080318
University of Minnesota MS/UMF Bridge grant B-0921-01
NSERC Postdoctoral Fellowship (CCD)
University of Minnesota MnDrive Postdoctoral Fellowship (CCD)

Title: Hippocampal Representations in rats worried about being attacked by a threatening robot in the “robogator” approach-avoidance task

Authors: *C. C. DAMPHOUSSE[1], O. L. CALVIN[1], M. T. ERICKSON[2], A. D. REDISH[1]; [1]Neurosci., Univ. of Minnesota, Minneapolis, MN; [2]Sch. of Med., Indiana Univ., Indianapolis, IN

Abstract: Computationally, “worry” requires an object of concern. An agent that is “worried” about an outcome must, somehow, represent that outcome. Hippocampal representations of non-local information are well-established in appetitive (approach) conditions, but have only been studied in very limited aversive (avoidance) conditions. In order to investigate the extent to which hippocampal representations dynamically represent situations to be approached or avoided, we trained rats on a task in which they were placed into conflict between approaching a food reward and avoiding a threatening robotic predator.

6 Brown-Norway rats (3M, 3F) were first trained to run back and forth along a closed linear track for alternating food rewards with a safe “nest” space at one end and a “far feeder” at the other. We next introduced a robot at the end of the maze opposite to the nest, closest to the far feeder. After 2 sessions of habituation in which the robot did not move, the robot would occasionally screech and surge forward as the rat ran from the nest to the far feeder. The robot did not attack for the first 15 laps, but then attacked randomly with a 20% chance, which typically induced a fight or flight response in the rat.
Once the animals began to be ‘attacked’ by the robot, the rats exhibited prospective fear by running fewer laps and moving more slowly down the track towards the robot. The slowed movement was characterized by a creeping behavior in which the animals alternately moved forward and paused before moving forward again. Rats also hesitated at the nest exit more frequently on attack days. We decoded hippocampal representations of outcomes during sharp-wave ripple (SWR) events during this hesitation, which revealed a shift away from the feeder to the robot after the animal was attacked.
In a separate experiment, we administered the anxiolytic drug diazepam (2 mg/kg) or Tween-20 (vehicle control) to 5 Brown-Norway rats (3M, 2F) on the same task. Diazepam significantly reduced both the number of SWR events and the prospective fear behavior of the animals. Hesitation was reduced and creeping behavior was absent under diazepam relative to vehicle control. These results suggest that worry may be mediated by hippocampal representations of the threatening robotic predator. The large reduction in SWR events while under the effects of diazepam suggests that the reduction in fear behavior may be due to disruptions in processing occurring during SWRs.

Disclosures: C.C. Damphousse: None. **O.L. Calvin:** None. **M.T. Erickson:** None. **A.D. Redish:** None.

Poster

PSTR168. Neural Mechanisms for Choice

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR168.21/OO7

Topic: H.03. Decision Making

Support: NSF 2234748

Title: Reaching Movements Reflect Ongoing Deliberation Prior to a Decision

Authors: *J. A. CALALO[1], T. T. NGO[1], S. R. SULLIVAN[1], A. R. ROTH[1], R. LOKESH[1], J. H. BUGGELN[1], K. STRAND[1], V. MARCHHART[1], M. J. CARTER[2], I. KURTZER[3], J. G. A. CASHABACK[1];
[1]Univ. of Delaware, Newark, DE; [2]McMaster Univ., Hamilton, ON, Canada; [3]New York Inst. of Technol., Old Westbury, NY

Abstract: We constantly make choices while moving, such as deciding whether to pass to the left or right of someone while walking down a hall. The few studies that examine the interplay between decision-making and movement have used a sudden target change (e.g., colour) to evoke an immediate decision and motor response, whereas perceptual decision-making research has manipulated goal-related evidence over time that influences the timing of a selected decision. In both cases, deliberation is hidden. Here we test the idea that decision-making and motor circuitry continuously interact during ongoing deliberation. We predicted that lateral hand movement would reflect ongoing deliberation, prior to a decision. We extended the well-known "tokens task" (Cisek, 2009) to require active forward movement prior to the final decision. Participants were required to move forward from a start position towards two potential targets. Once participants left the start position, 15 tokens individually moved in 160 ms increments into one of the two targets. Participants indicated the target expected to finish with the most tokens by both hitting the selected target with their reaching hand and pushing a button with the other hand. Interleaved with pseudo-random token patterns were biased token patterns, where the first three tokens moved into the left target (left bias) or right target (right bias). Critically, we measured the unconstrained lateral hand position, prior to the decision, to determine the influence of deliberation on movement. In Experiment 1, the left and right biased token patterns differentially impacted the lateral hand position prior to a decision ($p = 0.003$), supporting the idea that the ongoing deliberation is reflected in movements. In Experiment 2, we manipulated both the token bias direction and the rate of token movement. For both left and right bias token patterns, we moved the first four tokens either individually in 160 ms increments (slow rate) or all at once (fast rate). Again, we found that the left and right biased token patterns differentially impacted the lateral hand position prior to a decision ($p < 0.001$). However, participants made earlier decisions with a slow rate compared to a fast rate token pattern ($p < 0.001$), suggesting a temporal urgency component to the decision-making process. Our findings demonstrate that hand movements can reflect a continuous readout of the ongoing deliberation, supporting the idea that there is a continuous interaction between decision-making and motor circuitry.

Disclosures: J.A. Calalo: None. **T.T. Ngo:** None. **S.R. Sullivan:** None. **A.R. Roth:** None. **R. Lokesh:** None. **J.H. Buggeln:** None. **K. Strand:** None. **V. Marchhart:** None. **M.J. Carter:** None. **I. Kurtzer:** None. **J.G.A. Cashaback:** None.

Poster

PSTR168. Neural Mechanisms for Choice

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR168.22/OO8

Topic: H.03. Decision Making

Support: NIH Grant F30MH134507

Title: the neural mechanisms underlying categorical decision making

Authors: *N. STEINBERG[1], M. RIESENHUBER[2], K. A. ZAGHLOUL[3];
[2]Georgetown Univ. Med. Ctr., [1]Georgetown Univ. Med. Ctr., Washington, DC; [3]Natl. Inst. of Neurolog. Disorders and Stroke, NINDS, Bethesda, MD

Abstract: Our general knowledge of the meaning of words, images, concepts, and their associations in the world around us depends on bidirectional signaling: bringing in information from the world, in addition to accessing previous memories to make sense of it. The importance of integrating memory-driven information towards recognition is evident in several previous studies that show temporary inactivation of the anterior temporal lobe (ATL) leads to similar semantic deficits seen in semantic dementia (SD).Here, we leveraged intracranial electrocorticogram (iEEG) in epilepsy patients (n=26) to investigate the interaction between feedforward (perceptual) and feedback (memory-driven) signals to ultimately influence decision making. Prior to iEEG implantation, participants completed preoperative behavioral sessions where they viewed a series of both clear and ambiguous (blurred) images for 500 ms and reported which semantic category the stimuli belonged to: animal, object, person or place. Image blurriness was titrated to create roughly a 50% accuracy (while accuracy on their nonblurred counterparts approached ceiling). Participants then completed the same task during intracranial monitoring. Analyses on the behavioral data show that blurred stimuli were more difficult to categorize, as reflected by increased reaction time (RT) across all categories [$F(1, 25) = 44.5$, $p = 1.7 * 10^{-6}$]. We then analyzed the neural data to look for signals that corresponded to the increased RT for the blurred images with the high spatiotemporal precision offered by iEEG. Analyses suggest that regions of interest (ROIs) in the posterior ventral visual stream have early selectivity for nonblurred images but selectivity for their blurred counterparts occurs several hundred milliseconds later. However, the difference in selectivity latency across blur conditions decreases in a posterior-to-anterior fashion, including ATL, medial temporal lobe (MTL), and prefrontal cortex (PFC). This suggests that executive control and memory are recruited during times of relatively greater perceptual ambiguity, and is a requirement for correctly categorizing blurred images.

Disclosures: **N. Steinberg:** None. **M. Riesenhuber:** None. **K.A. Zaghloul:** None.

Poster

PSTR168. Neural Mechanisms for Choice

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR168.23/OO9

Topic: H.03. Decision Making

Support: Simons Collaboration on the Global Brain
McKnight Foundation
NIH RF1 NS132025
NIH R01 NS113110
NIH R01 NS131229
NIH R01 NS112312

Title: Tracking the emergence of persistent activity in premotor cortex across learning of motor planning task

Authors: *C. WANG[1], B. KANG[2], G. CHEN[1], S. DRUCKMANN[2], N. LI[1];
[1]Baylor Col. of Med., Houston, TX; [2]Stanford Univ., Palo Alto, CA

Abstract: Cognitive functions, such as short-term memory, are dependent on persistent neural activity that maintains information. Persistent activity is distributed across multiple brain regions, most prominently in frontal cortical areas. It remains unclear how persistent representations emerge over experience. In the mouse, neurons in the anterior lateral motor cortex (ALM) exhibit preparatory activity during motor planning, a form of persistent activity. We used longitudinal two-photon calcium imaging to track the emergence of preparatory activity in mice learning a motor planning task. Mice discriminate the position of a pole using their whiskers and report choice using a right or left lick. The sensory stimulus and motor response are separated in time by a delay epoch (3 s), so mice must use short-term memory to report the correct choice. Behaviorally, mice progressed from a naïve state to a trained state over a period of one month, as measured by the proportion of correct trials in a session (from 50% to above 70%). We deduced the mice's behavioral strategy using a latent state model. Specifically, a 3-state hidden Markov model with generalized linear model observations (GLM-HMM) was fit on behavioral data across different stages of training. At each stage of training, the GLM-HMM model discovered latent states that used task and behavioral variables to predict upcoming choice (Ashwood et al, 2022). Once fitted, the model's cross-validated accuracy for predicting the upcoming choice ranged from 45-70% for sessions from the naïve state and 60-90% for sessions from the trained state. In the naïve state, mice chose their lick direction based on the previous choice, previous pole position, or a unidirectional bias. On the other hand, in a trained state, the same mouse primarily used the current pole position to decide lick direction. ALM neurons exhibited activity that correlated with sensory stimulus (pole position), choice (lick direction), and trial outcome (correct vs. error). Across learning, we found that there was a significant increase in the proportion of choice-selective and outcome-selective neurons in the recorded ALM population (Chi-squared test, $X^2(1)=13.9$, $p=0.0002$; $X^2(1)=18.7$, $p=0.00002$, respectively). The increase in proportion of choice-selective neurons primarily occurred in the delay epoch and not in the sample and response epochs (Unpaired t-test, $p=0.93$, $p=0.000013$, $p=0.21$ for sample, delay and response epochs respectively). Taken together, these results outline a learning process that produces persistent neural representations to link past stimulus and future actions.

Disclosures: C. Wang: None. **B. Kang:** None. **G. Chen:** None. **S. Druckmann:** None. **N. Li:** None.

Poster

PSTR168. Neural Mechanisms for Choice

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR168.24/OO10

Topic: H.03. Decision Making

Title: Projection specific information coding in frontal cortical networks

Authors: *I. LENZI[1,2], A. DESPATIN[2], N. NOJAVAN LAHIJI[1,2], M. NEßELER[2], M. SPEHR[2], B. KAMPA[2], S. MUSALL[1,2];
[1]Forschungszentrum Jülich, Jülich, Germany; [2]RWTH Aachen Univ., Aachen, Germany

Abstract: The anterior lateral motor cortex (ALM) is crucial for integrating sensory information to drive associated behaviors. This process critically relies on long-range projections to subcortical regions, in particular the striatum which receives robust projections from cortico-striatal projecting neurons (CStr). A potential role for this prominent projection pathway is the formation of new sensorimotor associations when learning new behaviors. However, the specific role of CStr neurons during learning remains unclear. To address this issue, we anatomically characterized the distribution of ALM terminals in the striatum and found a gradient along the anterioposterior axis. This pattern was preserved for ipsi- versus contralateral projections, with contralateral being ~65% weaker compared to ipsilateral expression. Next, we retrogradely labeled CStr neurons in ALM and performed 2-photon imaging over a series of behavioral tasks with increasing cognitive demand. First, mice underwent an innate behavior, collecting a reward from two available water spouts, requiring no novel stimulus-response associations. Second, mice learned to associate the location of click sounds to the side of a water reward, and lastly had to retain their choice for a short delay period. Across sessions, we observed a clear increase in choice selectivity for all ALM neurons, starting with the introduction of the non-innate auditory task. CStr neurons were more selective as non-CStr neurons, although this effect was subtle compared to the overall learning-related changes. These changes persisted when repeating the innate task, with a 2-fold increase of choice selective neurons after learning, pointing to a general restructuring of choice-related circuitry. To causally confirm these results, we optogenetically inactivated either all or only CStr neurons in ALM during the innate and auditory tasks. Inhibiting CStr neurons during the innate task strongly impaired performance before, but not after training. To further isolate this effect, we used the synaptic silencer eOPN3 to specifically inactivate ALM projections to either the striatum, the thalamus or the superior colliculus (SC) during task learning. In agreement with our earlier results, we found that inhibiting CStr projections mostly reduced task performance early in training, whereas inhibiting projections to SC and thalamus became more relevant when further increasing cognitive demands. Overall, our results demonstrate that task learning induces long-lasting changes in choice-related circuitry in ALM with distinct subcortical projection pathways performing different roles in the acquisition of new behaviors.

Disclosures: I. Lenzi: None. **A. Despatin:** None. **N. Nojavan Lahiji:** None. **M. Neßeler:** None. **M. Spehr:** None. **B. Kampa:** None. **S. Musall:** None.

Poster

PSTR168. Neural Mechanisms for Choice

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR168.25/OO11

Topic: H.03. Decision Making

Support: NIH R01EY019041
NIH-NINDS U19NS107609-03
DOD VBFF
NIH T32GM007281
NIH F30EY033648

Title: The neural encoding and causal contributions of FEF, LIP, and SC to rapid categorical decisions

Authors: *O. ZHU[1], V. SHIRHATTI[1], K. LATIMER[1], O. GOZEL[2], S. DAVID[1], S. CHANG[1], A. MEDOFF[1], B. DOIRON[2], D. J. FREEDMAN[1], *O. ZHU[3];
[1]Neurobio., [2]Neurobio. and Statistics, [3]Univ. of Chicago, Chicago, IL

Abstract: Our ability to rapidly categorize incoming stimuli is an essential cognitive process that imparts meaning to incoming sensory stimuli and guides our behavior. Here, we designed a reaction-time, oculomotor, visual categorization task where the monkey is presented with a random dot motion stimulus and must saccade to a color target corresponding to a learned motion category. During task performance, we recorded populations of neurons using up to six linear microelectrode arrays (Plexon) simultaneously targeting the lateral intraparietal area (LIP), frontal eye field (FEF), and superior colliculus (SC) in two monkeys. On average, each session yielded around 50 well-isolated single units from each area. Using linear support vector machines, we found that population responses in each area reliably encoded task-related variables such as stimulus category, target configuration, and saccade direction prior to saccade onset. Overall, the strength and timing of encoding were similar between the three brain areas. To distinguish the contributions of LIP, FEF, and SC to rapid categorical decisions, we reversibly inactivated each area with muscimol while simultaneously recording the other two areas with linear arrays. We tested the monkey's behavior using two stimulus-target configurations such that either the motion stimulus or a saccade target was placed in the inactivated visual field (IF), to independently assess deficits in categorization or oculomotor control. When the motion stimulus was presented in the IF, we found significant behavioral deficits after inactivating each of the three areas, with the strongest accuracy deficits associated with FEF and SC inactivation. When a saccade target was presented in the IF, we observed significant behavioral deficits only during FEF and SC, but not LIP, inactivation. Consistent with these behavioral deficits, we also found a reduced magnitude and delayed latency of neuronal encoding of task variables in the population responses during inactivation. Our results suggest that FEF and SC play a causal role in both evaluating the visual stimulus and target selection during this rapid categorization task, whereas LIP is more engaged in the visual, rather than oculomotor, aspects of the categorization task. Ongoing analyses aim to quantify communication subspaces between simultaneously recorded areas to measure the magnitude and directionality of interactions between each area and whether these change after inactivation. We also use generalized multi-linear models (GMLM) to quantify how trial-to-trial variability in the low-dimensional dynamics of the populations in each area relates to variability in behavior.

Disclosures: O. Zhu: None. **V. Shirhatti:** None. **K. Latimer:** None. **O. Gozel:** None. **S. David:** None. **S. Chang:** None. **A. Medoff:** None. **B. Doiron:** None. **D.J. Freedman:** None. **O. Zhu:** None.

Poster

PSTR168. Neural Mechanisms for Choice

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR168.26/OO12

Topic: H.03. Decision Making

Title: Latent Task Variable Inference in an Audio-Visual 2-Alternative Forced Choice Contextual Decision-Making Task in Mice

Authors: *K. SAFARYAN[1], A. LAI[1], E. LI[1], G. ORTEGA[1], J. SHENASSA[1], P. GOLSHANI[2]; [1]UCLA, Los Angeles, CA; [2]UCLA Dept. of Neurol., Los Angeles, CA

Abstract: Cognitive flexibility allows animals to make rewarding decisions in rapidly changing environments. Flexible responses to the same stimulus in different contexts requires selective attention and continual appraisal of contextual variables. Yet, how distinct cortical circuits encode contextual variables and gate relevant and non-relevant stimuli to derive context-dependent decision making is not understood. To address this problem, mice were trained to perform serial extradimensional shifts, where cross-modal stimuli interchangeably define trial outcome or serve as distractor. Initially, mice were trained to perform a unimodal discrimination task where 45^{o} and 135^{o} visual gratings or high or low auditory tones signaled that the animal should lick the left or right lick ports, respectively. After 10-15 daily sessions, mice learned to perform the task at expert level, defined at performance of at least 85% correct trials for three consecutive training sessions. Mice were then trained to perform a compound discrimination task, where simultaneously presented auditory and visual stimuli served as either relevant or distractor signals. Finally, mice were trained to make serial extradimensional shifts, where learned to infer which stimulus modality was relevant and which stimulus modality the distractor. The shifts between relevant modalities were covert and accompanied by immediate fast decline of the performance with consequent gradual improvement. It took an additional 25-30 days for mice to learn to perform multiple switches back and forth during the same session. We estimated performance with parametric sigmoidal model fit to the performance in each block. Each block on average consisted of 90-100 trials with most frequent after-switch recovery period lasting 20-30 trials. We are currently recording from several cortical structures, including the primary visual cortex and anterior cingulate cortex, using Neuropixels probes to uncover the neural correlates of sensory and contextual variables.

Disclosures: K. Safaryan: None. **A. Lai:** None. **E. Li:** None. **G. Ortega:** None. **J. Shenassa:** None. **P. Golshani:** None.

Poster

PSTR168. Neural Mechanisms for Choice

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR168.27/OO13

Topic: H.03. Decision Making

Support: KAKENHI 19H03531
KAKENHI 21K15608

Title: Synchronous gamma-band oscillation dynamically changes between sensory and decision related areas during flexible decision making

Authors: *Y. SUDA, T. UKA;
Dept. of Integrative Physiology, Grad. Sch. of Med., Univ. of Yamanashi, Chuo, Japan

Abstract: Flexible decision making is an indispensable ability for humans. Neurons in the lateral intraparietal area (LIP) accumulate relevant information preferentially depending on context, but, how sensory information represented in visual cortex, area MT, is conveyed to stages of evidence accumulation remains unclear. To elucidate this question, we investigated the functional connectivity between MT and LIP by recording simultaneously from both areas using two electrodes while monkeys performed a switching task. Two Japanese macaques were trained to flexibly switch between a direction discrimination task and a depth discrimination task. Difficulty of the tasks was varied by changing the percentage of coherently moving and binocularly correlated dots in a random dot stimulus. We recorded local field potential (LFP) in areas MT and LIP simultaneously with a single electrode. The preferred choice target was placed toward the response field of the LIP neuron, whereas the null choice target was positioned diametrically opposite to the preferred choice target. The random dot stimulus optimized to preference of MT neurons was presented in the receptive field of that neurons. To examine whether the oscillation changed depending on pairs connection between MT and LIP, we grouped the MT-LIP pairs by task relevance into two groups, “Task congruent pairs” and “Task incongruent pairs”. Whereas the former pair was task relevant in which preference of MT neurons coincided with the response filed of LIP neurons for task requirement, the latter was task irrelevant in which pairs connection was incongruent for making a correct decision. To examine the functional connectivity, we calculated pairwise phase consistency (PPC) between the two electrodes. We confirmed that the gamma-band oscillation of task congruent pairs was significantly larger than that of task incongruent pairs during decision formation. Granger causality analysis demonstrated that the gamma-band oscillation had a direction from area MT to LIP, suggesting that the gamma-band synchrony might be bottom-up signal related to context dependent decision making. In incongruent MT neurons, where task relevant connection changed depending on indicated task, the enhancement of gamma oscillation occurred only in case of MT-LIP connection congruent trials. These results suggest that the gamma-band bottom-up oscillation from area MT to LIP might be related to enhance task relevant connection according to task requirement for flexible decision making.

Disclosures: Y. Suda: None. **T. Uka:** None.

Poster

PSTR168. Neural Mechanisms for Choice

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR168.28/OO14

Topic: H.03. Decision Making

Support: NIH Grant R01 MH132732

Title: Lc-ne system improves performance in a visual evidence accumulation task

Authors: *H. XIA[1], C. ROSE[1], G. KANE[1], S. LI[1], B. B. SCOTT[2];
[2]Psychological & Brain Sci., [1]Boston Univ., Boston, MA

Abstract: Norepinephrine (NE) has been shown to play a significant role in sensory processing. However, the precise function of NE in perceptual decision making remains elusive. We employed a free-response evidence accumulation task, where rats chose a reward port based on a series of randomly timed 10ms light flashes from left and right LEDs. Rats received rewards for choosing the port that associated with a greater probability of light pulses. In order to evaluate the effect of NE on this task, we administered clonidine, an agonist of α2-adrenergic receptors, into six adult female Long-Evans rats (5, 10, and 20 µg /kg I.P.). Clonidine administration increased both accuracy and reaction time (RT) in a dose-dependent manner. To control for non-specific targets of clonidine, we selectively transduced LC-NE neurons with CAV-PRS-hm3Dq-mCherry to activate LC-NE cells during behaviors in both male and female rats (n=6). Similar to the administration of clonidine, the injection of DREADD agonist deschloroclozapine (DCZ, 0.5mg/kg, i.p.) increased RT and accuracy compared with saline controls. Behavioral analysis indicated that rats solve this task by integrating light pulses from both sides and subsequently selecting a side once they reach an internal threshold. To further assess the impacts of NE manipulations on accumulation behavior, we fitted the drift diffusion model to data from over 120,000 trials, which describes two alternative forced choice as a noisy accumulation process approaching a decision boundary. Rats that received clonidine injections exhibited a wider separation in the decision boundary, indicating that they require additional evidence in order to meet criteria for making decisions. Rats injected with excitatory DREADD showed similar fitting results following DCZ injections. Together these results suggest a role for the noradrenergic system in regulating evidence accumulation during perceptual decision making.

Disclosures: H. Xia: None. **C. Rose:** None. **G. Kane:** None. **S. Li:** None. **B.B. Scott:** None.

Poster

PSTR168. Neural Mechanisms for Choice

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR168.29/OO15

Topic: H.03. Decision Making

Support: The French Ministry of Higher Education and Research Grant

Title: Using behavioral and computational analyses to predict subtypes of apathy in an effortful cost/benefit task.

Authors: *E. E. BRADLEY[1], G. PAGNIER[1], U. TISSOT[2], A. BONNEFOND[2], G. LAFOND-BRINA[2];
[1]Neurosci., Brown Univ., Providence, RI; [2]French Natl. Inst. of Hlth. and Med. Res., Univ. of Strasbourg, Unity 1114 - INSERM, Paris, France

Abstract: Apathy is a common behavioral disorder exhibited in about 40% of Parkinson's Disease patients and is associated with a reduced ability to initiate goal-oriented movements and a decreased quality of life. Computational models suggest apathy may emerge from multiple possible circuits rooted in the basal ganglia. Thus, identifying behavioral signatures accompanying specific subtypes of apathy may help guide individual treatment. Here, we assess data collected from 91 patients with clinical apathy stratified into the following subtypes as determined from the Dimensional Apathy Scale: 1) emotional, 2) initiative, 3) executive, 4) control. Participants completed 180 trials of an effortful decision-making task designed to orthogonally quantify participants' sensitivity to reward and mental effort. We systematically varied effort and reward levels to produce four different types of trials: effort (holding reward between the two options constant), reward (holding required effort levels constant), optimality (a high effort, low reward option versus a low effort, high reward option) and preference (a high effort, high reward option versus a low effort, low reward option). We present trial by trial analyses showing that participants' accuracy and reaction times are modulated by subtype apathy. Specifically, linear mixed effects modeling suggests participants diagnosed with initiative apathy exhibit deficits in choosing the optimal option (t=-1.515). In secondary analyses, we investigated the within-subject effect of fatigue on trial accuracy and found that apathy subtype modulates lapse rate in a trial-specific way. To integrate both behavioral choices and participants' reaction times we present complimentary drift diffusion models highlighting group differences between apathy subtypes. We argue that these computational and behavioral results capture key differences between apathy subtypes and highlight the capability of using non-invasive behavioral measures as diagnostic tools.

Disclosures: E.E. Bradley: None. **G. Pagnier:** None. **U. Tissot:** None. **A. Bonnefond:** None. **G. Lafond-Brina:** None.

Poster

PSTR168. Neural Mechanisms for Choice

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR168.30/OO16

Topic: H.03. Decision Making

Support: SNSF Early PostdocMobility P2BEP3_200212

Title: Neural activity in anterior cingulate cortex reflects movements and task variables

Authors: *L. T. OESCH, D. SANDBERG, J. COUTO, A. K. CHURCHLAND;
Neurobio., UCLA, Los Angeles, CA

Abstract: Recent work shows that movements modulate neural activity broadly in mice performing perceptual decision-making tasks. Movement signals are frequently better predictors of neural activity than task variables even in primary sensory regions. However, it is unknown whether movement signals similarly dominate neural activity in brain regions traditionally thought of as more "cognitive". Here, we trained freely moving mice on a visual or auditory rate accumulation task and recorded the activity of excitatory neurons in the anterior cingulate cortex (ACC) using miniaturized fluorescence microscopes. We estimated the neural variance explained by task- and movement variables by fitting linear encoding models to the neural data. We included task variables, such as current/previous choice and outcome alongside movement variables, such as head orientation angles, the position of a set of body parts and components of the behavioral video. In keeping with prior results, movements accounted for considerable variance in neural activity. Surprisingly, task variables likewise explained considerable variance, more so than was present in previous work, and they further accounted for a large amount of uniquely explained variance. Importantly, the unique explained variance of the task variables was positively correlated with the total amount of variance for each neuron whereas there was no such correlation for the video components. Finally, we found that task variables made strong unique contributions throughout the trial, including the stimulus presentation period that is devoid of instructed movements. Interestingly, the unique contribution of task variables was elevated just before trial initiation, reflecting prominent encoding of previous choice and outcome. Taken together, these findings suggest that excitatory ACC neurons strongly encode not only movement variables, but task variables as well, in contrast with other areas. This argues that movement signals, though clearly present in cognitive areas, might sometimes take a back seat to cognitive signals.

Disclosures: L.T. Oesch: None. **D. Sandberg:** None. **J. Couto:** None. **A.K. Churchland:** None.

Poster

PSTR169. Distributed Mechanisms of Working Memory and Decision-Making I

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR169.01/OO17

Topic: H.05. Working Memory

Support: Bordeaux Neurocampus junior chair program, co-funded by Région Nouvelle-Aquitaine and the University of Bordeaux Initiative of Excellence (IdEx) (F.B.W. Neurocampus junior chair)
European Union's Horizon Europe research and innovation program under

European Research Council (ERC) Starting Grant agreement N° 101040391 (F.B.W. MEMOPROSTHETICS)

Title: Combined neural recordings and stimulation of hippocampal and cortical structures in macaques performing a visuospatial memory task

Authors: *A. GUPTA[1], A. BOISSENIN[3], N. VARDALAKIS[3], M. TAILLADE[2], H. ORIGNAC[1], T. NGUYEN[1], A. SADOUN[4], F. B. WAGNER[5];
[1]Univ. of Bordeaux, Bordeaux, France; [2]Univ. of Bordeaux, Bordea, France; [3]Univ. de Bordeaux, Bordeaux, France; [4]Motac, Bordeaux, France; [5]Ctr. Natl. de la Recherche Scientifique, Bordeaux, France

Abstract: Efficient memory encoding and retrieval are correlated with neural oscillations in the theta (4-8 Hz) and gamma (30-80 Hz) ranges across distributed hippocampo-cortical networks. These are affected by a wide range of neurological disorders. Recently, Deep Brain Stimulation (DBS) of limbic structures has been investigated as a potential treatment for memory disorders such as Alzheimer's disease. Yet, it remains unknown how DBS affects hippocampal and cortical theta and gamma oscillations in memory tasks. We aimed to investigate this question in non-human primates trained to perform a visuospatial paired associative learning task.
To this end, we designed a new implant consisting of two DBS electrodes with 8 contacts each, to target respectively the hippocampus (along its anteroposterior axis) and entorhinal cortex, as well as an electrocorticographic (ECoG) grid with 60 contacts spanning the prefrontal and premotor cortices. These neural implants had dimensions tailored to the macaque brain and were integrated into a single percutaneous titanium connector. When interfaced with specialized external hardware for combined neurophysiological recordings and multisite electrical stimulation (Blackrock Neurotech, USA), this unique setup allows us to simultaneously record field potentials and stimulate on all 76 contacts.
We implanted this distributed electrode assembly in a 12-year-old macaque monkey using a neuronavigation system based on Magnetic Resonance Imaging. We then recorded epicortical and local field potentials from all 76 electrode contacts when the animal performs a visuospatial paired associates learning task in freely behaving conditions. Time-frequency analysis of ECoG signals revealed task-related activity during memory encoding and retrieval. Electrical stimulation delivered in the passive state (single biphasic pulses, 300-1000 μs pulse width, 0.1-10 mA) elicited evoked responses across the ECoG and depth electrodes, showing our ability to target the hippocampo-cortical network at a network level. Future experiments will determine the effects of electrical stimulation on neural oscillations and cognitive performance during the paired associated learning task.

Disclosures: A. Gupta: None. **A. Boissenin:** None. **N. Vardalakis:** None. **M. Taillade:** None. **H. Orignac:** None. **T. Nguyen:** None. **A. Sadoun:** None. **F.B. Wagner:** None.

Poster

PSTR169. Distributed Mechanisms of Working Memory and Decision-Making I

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR169.02/OO18

Topic: H.05. Working Memory

Support: STI2030_Major Project(2021ZD0203600)

Title: Theta and gamma oscillations in the prefrontal-hippocampal loops support sequential working memory

Authors: ***M. SU**[1,2,3], K. HU[4], B. SUN[4], S. ZHAN[4], Z. YE[5];
[1]CEBSIT, Shanghai, China; [2]Inst. of Psychology, Chinese Acad. of Sci., Beijing, China; [3]Univ. of Chinese Acad. of Sci., Beijing, China; [4]Dept. of Neurosurgery, Ctr. for Functional Neurosurgery, Ruijin Hospital, Shanghai Jiao Tong Univ. Sch. of Medicine,, Shanghai, China; [5]Inst. of Neuroscience, Ctr. for Excellence in Brain Sci. and Intelligence Technology, Chinese Acad. of Sciences,, Shanghai, China

Abstract: The dorsolateral prefrontal cortex (DLPFC) and hippocampus are engaged for processing visual features of a single object and sequential relations of multiple objectsonline (sequential working memory). However, it remains unclear how the DLPFC interacts with the hippocampus in sequential working memory. We addressed this issueusing stereoelectroencephalography (SEEG) with an original line ordering task intwenty patients with epilepsy (eight women, age 27.0±8.1 years). Participants showed longer thinking times and more recall errors when asked to arrange random lines clockwise (random trials) than to maintain ordered lines (ordered trials) before recalling the orientation of a particular line. SEEG recordings were obtained from 43 sites in the DLPFC and 56 sites in the hippocampus. First, the ordering-related increase in thinking time and recall error was associated with a transient theta power increase in the hippocampus and a sustained theta power increase in the DLPFC (3-10 Hz). The hippocampal (but not DLPFC) theta power increase correlated with the memory precision of line orientation. Second, the time-resolved gamma activity (55-95 Hz) in the DLPFC represented the serial order, and the time-averaged gamma activity in the hippocampus correlated with the memory precision of line orientation. Third, theta phase coherences between the DLPFC and hippocampus were enhanced for ordering, especially for more precisely memorized lines. Fourth, the theta band DLPFC→hippocampus influence was selectively enhanced for ordering, especially for more precisely memorized lines. This study suggests that in the DLPFC-hippocampal loops, both theta and gamma oscillations support sequential working memory, but in different manners. Theta oscillations may mediate the DLPFC-hippocampal interactions for the online processing of sequential relations among objects. Gamma oscillations may represent the serial order in the DLPFC and the visual features in the hippocampus.

Disclosures: **M. Su:** None. **K. Hu:** None. **B. Sun:** None. **S. Zhan:** None. **Z. Ye:** None.

Poster

PSTR169. Distributed Mechanisms of Working Memory and Decision-Making I

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR169.03/OO19

Topic: H.05. Working Memory

Support: STI2030-Major Projects, No.2021ZD0203700/2021ZD0203705
NYU Shanghai Summer Undergraduate Research Program (SURP)

Title: Cardinal repulsion in working memory requires sensory-memory network interactions

Authors: J. YANG[1], **H. ZHANG**[2,3], *S. LIM[4,2];
[1]Weiyang College, Tsinghua Univ., Beijing, China; [2]Shanghai Frontiers Sci. Ctr. of Artificial Intelligence and Deep Learning, [3]New York Univ. Shanghai, Shanghai, China; [4]New York Univ. Shanghai, Shanghai city, China

Abstract: Exaggerating a nearly vertical or horizontal line as more tilted away from the cardinal axes, known as cardinal repulsion has been reported in many perceptual tasks. Recent works on visual working memory suggested that cardinal repulsion increases during memory after the orientation stimulus disappears, evidence for drift dynamics toward oblique orientations. Also, the variance of working memory reports for different orientations shows an uneven pattern, reaching minima at the cardinals and maxima at the obliques. Such bias and variance patterns are inconsistent with traditional working memory models, which assume all orientations are equally represented and maintained. With strong recurrent connections homogeneous for all orientations, traditional models predict no drift dynamics and a uniform variance pattern.// To understand the circuit mechanisms of visual working memory that produce bias and variance patterns observed experimentally, we adapted the neural code developed to account for cardinal repulsion during perception, that is, efficient coding combined with Bayesian perception originally suggested for sensory circuits. We first implemented such code in memory circuits where recurrent connections are strong enough to support persistent activity and heterogeneous to create uneven dynamics for different orientations. With attractors formed at oblique orientations, the models can reproduce the correct bias and variance patterns. However, they typically generate excessively fast drift, leading to rapid forgetting of the exact orientation during working memory. Also, fine-tuning the degree of heterogeneity is required, which can be easily disrupted by perturbations in network connectivity.// On the other hand, two interacting modular networks, sensory circuits with heterogeneous recurrent connections and memory circuits with homogeneous ones as the traditional working memory models, can reproduce the bias and variance patterns more robustly. The drift speed can be better regulated in the sensory-and-memory interacting networks. Furthermore, we found that stronger inhibitory tuning at the cardinal orientations is required in the sensory circuits to correctly shape bias and variance patterns. Overall, our work provides novel insights into the role of interactions between sensory and memory circuits through the lens of bias and variance patterns of working memory.

Disclosures: J. Yang: None. **H. Zhang:** None. **S. Lim:** None.

Poster

PSTR169. Distributed Mechanisms of Working Memory and Decision-Making I

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR169.04/OO20

Topic: H.05. Working Memory

Support: NIH Brain Initiative Grant RF1MH124909
BBRF Young Investigator Grant 30064

Title: The effects of beta rhythm perturbation on executive functions in epilepsy patients

Authors: *I. ALEKSEICHUK[1], T. A. BERGER[2], S. KOENIG[3], M. WISCHNEWSKI[2], C. SAIOTE[1], J. A. PEBBLES[1], R. A. MCGOVERN[4], M. C. PARK[4], A. B. HERMAN[5], D. P. DARROW[4], A. OPITZ[2];
[2]Biomed. Engin., [3]Psychiatry and Neurosurg., [4]Neurosurg., [5]Psychiatry, [1]Univ. of Minnesota, Minneapolis, MN

Abstract: Synchronization and desynchronization of the brain oscillations reflect and reciprocally control various brain functions, such as those underlying executive control in humans. Recently it was shown using invasive recordings that transcranial alternating current stimulation (tACS) can modulate frequency-specific brain oscillations, inducing phase entrainment of the neural spikes at relatively higher intensities and perturbation of intrinsic spike-field coherence at lower intensities. Both effects present great interest from scientific and clinical perspectives. In executive function and working memory, both theta synchronization and beta desynchronization are known correlates of successful cognitive acts. Here, we investigated the mechanisms of lower-intensity tACS in the theta and beta range and its effects on working memory performance by leveraging invasive electroencephalography in epilepsy patients. Six neurosurgical patients who met established clinical guidelines and voluntarily agreed to participate were implanted with subdural grids and depth electrodes (~100-200 contacts per patient) based on their medical requirements. The study was conducted in accordance with the Declaration of Helsinki and IRB regulations (University of Minnesota). Participants performed an N-back working memory task before and during beta, theta, or sham modulation at the bilateral temporal cortex in a crossover study design. The generalized linear mixed-effect model reveals a significant effect of stimulation on memory performance ($F = 15.5$, $p < 0.001$) but not on the reaction time ($p > 0.1$). Post-hoc t-tests show improvement ($p < 0.05$) following beta tACS compared to sham or theta modulation, while the latter induced no changes. Within-subject Hierarchical Drift Diffusion Model using Bayesian estimation shows a significant effect of beta stimulation relative to theta stimulation on the evidence accumulation process (drift rate) both at the group level (Bayes factor, $BF > 10$) and at single subject level ($BF > 3$ for every patient). Further, we found a significant perturbation of the beta oscillations at the temporal cortex during low-intensity beta tACS, which correlated with the cognitive effects ($p < 0.05$). Together, these results provide causal evidence for the role of temporal beta oscillations in the working memory in humans and highlight this activity as a candidate for cognitive therapy in epilepsy patients.

Disclosures: I. Alekseichuk: None. **T.A. Berger:** None. **S. Koenig:** None. **M. Wischnewski:** None. **C. Saiote:** None. **J.A. Pebbles:** None. **R.A. McGovern:** None. **M.C. Park:** None. **A.B. Herman:** None. **D.P. Darrow:** None. **A. Opitz:** None.

Poster

PSTR169. Distributed Mechanisms of Working Memory and Decision-Making I

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR169.05/OO21

Topic: H.05. Working Memory

Title: Eeg measure of bias in contralateral processing during visual short-term memory

Authors: *S. SHEREMATA[1], V. WISEMAN[2];
[2]Neurosci. Grad. Program, [1]Florida Atlantic Univ., Boca Raton, FL

Abstract: In the human brain, measures of visual short-term memory (VSTM) show a contralateral bias. Remembering objects results in greater activity in the contralateral as compared to the ipsilateral hemisphere. fMRI studies have shown that this contralateral bias is stronger in the left as compared to the right hemisphere. It has been suggested these hemispheric asymmetries reflect attentional processing demands inherent in working memory tasks. Specifically, as attention demands increase, the right hemisphere redirects attention resources across the visual field. However, differences in BOLD signal reflect the sum of each cognitive component involved in a task, limiting its utility for contrasting cognitive components. In contrast, electroencephalography (EEG) measures have been tied to specific cognitive functions. Specifically, alpha (8-12 Hz) power is suppressed contralateral to the focus of spatial attention while the contralateral delay activity (CDA) increases with each additional remembered item in the contralateral visual field. Therefore, EEG measures are ideal for differentiating between attention and working memory contributions to hemispheric asymmetries. To investigate the relative contributions of spatial attention and VSTM processes, an analysis of contralateral bias was performed on a publicly available data set (Gunseli et al., 2019). Response to items in the ipsilateral visual field was expected to be greater for right vs. left scalp locations. Because both alpha suppression and the CDA are contralateral measures (contralateral-ipsilateral), we predicted decreased measures of both alpha suppression and the CDA for right, as compared to left, scalp locations. The results demonstrate asymmetries in both measures, suggesting that the right hemisphere processes information across the visual field during attention and VSTM.

Disclosures: S. Sheremata: None. **V. Wiseman:** None.

Poster

PSTR169. Distributed Mechanisms of Working Memory and Decision-Making I

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR169.06/OO22

Topic: H.05. Working Memory

Title: Temporal regularities facilitate auditory working memory via motor-sensory interplay

Authors: *S. TIAN, H. LUO;
Peking Univ., Beijing, China

Abstract: Temporal regularities are characteristics of natural auditory experiences such as speech and music. They are known to facilitate auditory perception, yet their contribution to auditory working memory (WM) and the underlying neural basis remain unclear. Here we performed an auditory WM experiment combined with electroencephalography (EEG) recordings and computational modeling to address the question. Human subjects were instructed to temporarily retain in WM a sequence of 12 piano tones presented rhythmically or arrhythmically. After the maintaining period, a probe tone sequence with random inter-tone interval was presented, and subjects reported whether or not they were identical to the preceding sequence in pitch. Behaviorally, although having comparable accuracy, rhythmic tone sequence showed faster response time (RT) than the arrhythmic condition, and hierarchical drift-diffusion model fitting reveals that the decreased RT for rhythmic sequence arises from lower response boundary in perceptual decision-making. Furthermore, during sequence presentation (encoding), rhythmic sequence elicits enhanced phase-locked theta- and alpha-band (3-11 Hz) responses and stronger non-phase-locked beta-band responses in the frontoparietal region. During WM retention, the rhythmic condition displays stronger theta-band activations, implying better memory maintenance. Most importantly, the non-phase-locked beta-band power during encoding, a neural index of motor engagement, correlates with subsequent theta-band power during maintenance and also predicts lower response boundaries in computational model fitting. This implicates that temporal regularities drive the motor system to enhance predictive timing (via beta-band), which leads to efficient attention distribution over tone sequences and better memory maintenance. Taken together, temporal regularities facilitate auditory WM through intricate oscillation-based interactions between the motor and auditory systems.

Disclosures: S. Tian: None. **H. Luo:** None.

Poster

PSTR169. Distributed Mechanisms of Working Memory and Decision-Making I

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR169.07/OO23

Topic: H.05. Working Memory

Support: NRF Grant 2021M3E5D2A01019538

Title: The connectivity dynamics of working memory sub-process in the frontal-parietal-medial temporal networks

Authors: *D. KIM[1], B. LEE[2], J. KIM[3], C. CHUNG[4];
[1]Seoul Natl. Univ., Seoul, Korea, Republic of; [2]Dept. of Brain and Cognitive Sciences, Colleg, Looxid Labs, Seoul, Korea, Republic of; [3]Seoul Nat Univ., Seoul, Korea, Republic of; [4]Neurosurg., Seoul Natl. Univesity, Seoul, Korea, Republic of

Abstract: Introduction: For a long time, dominant views of working memory have emphasized the key role of the prefrontal cortex. However, recent studies found that the medial temporal lobe (MTL) and posterior parietal regions interact with prefrontal lobe, respectively, to perform working memory. Until now, it is not well known how these three regions interact when performing WM. Here, by analyzing neural signals during a working memory task, we address the gap in understanding the exact mechanism by which the frontoparietal network and MTL coordinate cognitive functions to control working memory.**Methods:** We recorded electrocorticography (ECoG) directly from frontal, medial temporal and parietal sites in 16 adults while they performed a visuospatial working memory task that is known to recruit the frontal, medial temporal and parietal regions. Partial directed coherence (PDC) was used for the connectivity analysis. Bidirectional PDC change value compared to baseline between the ECoG channels (frontal, parietal, and MTL) at distinct frequency bands, theta (3-7 Hz), alpha (8-13 Hz), beta (14-30 Hz), and gamma (31-120 Hz), were calculated.**Results:** ECoG signals were recorded from frontal (n=200), parietal (n=106) and MTL (n=31). In the maintenance phase, alpha connectivity from frontal and parietal to MTL was decreased compared to baseline. Gamma connectivity from parietal to frontal area was increased compared to baseline. In the operation phase, alpha connectivity from frontal to parietal area was decreased compared to baseline. Beta connectivity from parietal to frontal area was decreased compared to baseline, but from parietal to MTL was increased compared to baseline. **Conclusions:** We confirmed the connectivity dynamics of working memory sub-process in the frontal-parietal-MTL networks. In the maintenance phase, information should be held over a short period of time in the absence of the stimuli. It can be supposed that alpha connection with other regions decreases while information is maintained in the MTL. Meanwhile, gamma-band oscillation in the frontoparietal regions may be involved in the active maintenance of working memory information. In the operation phase, subjects should manipulate the information according to the task-specific operational cues. The frontal region continues to operate as a central executive, which may result in a decrease in both alpha and beta frontal lobe connections. On the other hand, goal-oriented working memory may be supported by interactions between MTL and parietal lobe in beta band.

Disclosures: D. Kim: None. **B. Lee:** None. **J. Kim:** None. **C. Chung:** None.

Poster

PSTR169. Distributed Mechanisms of Working Memory and Decision-Making I

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR169.08/OO24

Topic: H.05. Working Memory

Support: NSFC Grant 31730038

Title: The neural substrates of general cognitive ability based on multiple cognitive tasks

Authors: *L. ZHANG[1], G. XUE[2];
[1]BeijingNormal Univ., Beijing, China; [2]State Key Lab. of Cognitive Neurosci. and Learning, China, China

Abstract: Intelligence (IQ) is typically defined as a general cognitive ability (GCA) and challenges on its measurement and neural substrates remain. Specifically, many studies used Raven's Matrices (Raven) or working memory such as N-back task as a surrogate of IQ, whereas other studies extracted the g factor from multiple cognitive tasks. We wondered how the number of tasks affected the GCA measurement and the neural substrates of GCA measured from multiple cognitive tasks. We collected behavioral data on 20 cognitive tasks (including Raven) from 1760 participants and neural data (resting and task-state) from 745 of them. To benchmark the influence of tasks number on the reliability of g factor scores extraction, we extracted pairs of g factor scores (GCA) by sampling non-overlap 3-10 pairs of task indicators (20 in total) from the 19 non-Raven tasks. Results showed that pairs of GCA extracted from more tasks had better correlations between each other. A larger range (3-18) of task indicators sampling showed GCA extracted from more tasks had larger correlation with Raven scores. We then feed these GCA scores into connectome-based predictive modeling (CPM) to predict them from brain functional connectivity (FC) patterns. Similarly, as the number of task indicators used increased, there was a corresponding increase in the overlaps between pairs of model-selected networks that predicted the GCA and the explained variance by the CPM models. With these results discovered, we sticked to the GCA extracted from all the task indicators to delineate the neural correlates of GCA. We found that GCA was better predicted by task-state brain FC patterns than resting-state and GCA had a higher association with brain than Raven. Further analysis revealed that a wide variety of brain networks were selected by CPM models indicating GCA is supported by distributed networks. All these results suggest that GCA extracted by multiple tasks can be reliably utilized as a measure of IQ and the psychometric properties of GCA can be enhanced by adding more tasks regardless of the cognitive domain. In addition, as GCA surpassed Raven in measuring IQ, future studies should include more cognitive tasks when exploring the neural substrates of IQ.

Disclosures: L. Zhang: None. **G. Xue:** None.

Poster

PSTR169. Distributed Mechanisms of Working Memory and Decision-Making I

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR169.09/OO25

Topic: H.05. Working Memory

Support: NIH grant K23 AG072030
NIH grant K12 NS080223
NIH grant R01 EY017077

Title: Human oscillatory LFP activity during visual working memory

Authors: B. SINGH, Z. WANG, L. M. MADIAH, S. E. GATTI, J. N. FULTON, G. W. JOHNSON, D. J. ENGLOT, S. K. BICK, S. WILLIAMS ROBERSON, ***C. CONSTANTINIDIS**;
Vanderbilt Univ., Nashville, TN

Abstract: A distributed network of brain areas is activated during the performance of working memory tasks. Oscillatory activity is thought to be a marker of cognitive processes, although its role and distribution across the brain has been a matter of debate. To understand how oscillatory activity differentiates tasks and brain areas we collected local field potentials (LFPs) from intracranial depth electrodes implanted in 12 epilepsy patients to localize medically refractory seizures. The patients performed visual-spatial and shape working memory tasks with variable delay periods while LFPs were recorded. Data were acquired at 512 Hz sampling rate and were band-passed filtered between 0.5-200 Hz. We calculated induced LFP power using a multi-taper method. We then examined LFP power around task events after subtracting baseline power at each frequency, computed in the intra-trial interval. Power recorded in this manner revealed an extensive network of cortical and subcortical regions that were activated during the presentation of the visual stimuli and during their maintenance in working memory, including occipital, parietal, temporal, insular, and prefrontal cortical areas, and subcortical structures including the amygdala and hippocampus. Across most brain areas, the appearance of a stimulus produced broadband power increase; gamma power was evident during the delay interval of the working memory task. Differences between areas were also observed. Occipital cortex was characterized by elevated power in the high gamma (100-150 Hz) range during the 500 ms of visual stimulus presentation, which was less pronounced, or absent in other areas. The difference in power was significant across brain regions (1-way ANOVA, p=3.9e-18). A decrease in power centered in beta frequency (16-40 Hz) was also observed after the stimulus presentation, whose magnitude differed across areas (1-way ANOVA, p=5.7e-5), as did its relative timing. It reached a minimum at the occipital cortex after the stimulus offset, at the parietal cortex 150 ms later, and the prefrontal cortex another 650 ms later. These results reveal the interplay of oscillatory activity across a broad network of areas and region-specific signatures of oscillatory processes associated with visual working memory.

Disclosures: B. Singh: None. **Z. Wang:** None. **L.M. Madiah:** None. **S.E. Gatti:** None. **J.N. Fulton:** None. **G.W. Johnson:** None. **D.J. Englot:** None. **S.K. Bick:** None. **S. Williams Roberson:** None. **C. Constantinidis:** None.

Poster

PSTR169. Distributed Mechanisms of Working Memory and Decision-Making I

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR169.10/PP1

Topic: H.05. Working Memory

Support: NIH/NIMH Grant R01 MH128278 (JED)

Title: Faster response times in a verbal working memory task are associated with increased left crus I connectivity with the left central executive network in the post-task resting state

Authors: *A. S. COTTON[1], Y.-S. SHEU[2], J. E. DESMOND[3];
[1]Neurol., Johns Hopkins Univ., Baltimore, MD; [2]Dept. of Neurology, Cognitive Neurosci. Div., Johns Hopkins Sch. of Med., Baltimore, MD; [3]Dept. of Neurol. Div. of Cognitive Neurosci., The Johns Hopkins Univ. Sch. of Med., Baltimore, MD

Abstract: The cerebellum has been proposed to coordinate the sequential progression of verbal working memory (VWM) items in the phonological loop in a manner similar to how it facilitates the execution of motor sequences. An important component of the circuity that supports VWM is the central executive network (CEN). Task-based fMRI studies suggest that the CEN includes brain regions such as the dorsolateral prefrontal and inferior parietal cortex, as well as the lateral cerebellum. CEN areas can also be isolated through analyses that identify resting-state networks. Building on a previously published study, we used an independent component analysis (ICA) to analyze resting-state data collected after a VWM task and examine the hypothesis that mean trial response times (RTs) are associated with differences in left CEN connectivity in the cerebellum. A total of 19 subjects (age range=19-30; F=12, M=7) were recruited to the Johns Hopkins Medical Center and underwent fMRI scanning on a 3T Philips Achieva MRI Scanner. They completed 2 runs of a VWM task, during which they were instructed to memorize a sequence of 6 letters, recall the first several letters, and then specify whether or not a presented letter would occur next in the sequence. Afterwards, subjects underwent ~10 minute resting-state scans. For each subject, RTs were calculated and averaged for all trials across the 2 VWM task runs. Resting-state scans were analyzed in the CONN toolbox and preprocessed using the default pipeline. Preprocessed scans were entered into a 25 component ICA. The left CEN was among the resting-state networks identified. Through dual regression, subject-level CEN maps were calculated and entered into a correlation analysis with the mean VWM trial RTs. These were negatively correlated with left CEN connectivity to left crus I (Peak T(17)=-6.85; Peak MNI Corr.= -48,-48,-36; k=242) for a cluster threshold of $p<0.05$ and a voxel threshold of $p<0.001$. Our correlation suggests that faster RTs for the VWM task trials are associated with greater integration of left crus I into the left CEN. This correlation was contralateral to the intrinsic cerebellar connectivity associated with the left CEN but corresponded with activation that was related to faster RTs in an auditory n-back task. Our result provides evidence that such findings are related to greater cerebellar coordination with regions involved in VWM and are not merely artifacts of motor coordination. Unfortunately, our analysis cannot determine whether the connectivity patterns were preexisting for subjects with faster RTs or whether the connectivity patterns arose as a result of the task itself. We hope to examine this question in the future.

Disclosures: A.S. Cotton: None. **Y. Sheu:** None. **J.E. Desmond:** None.

Poster

PSTR169. Distributed Mechanisms of Working Memory and Decision-Making I

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR169.11/PP2

Topic: H.05. Working Memory

Support: NIH Grant R15MH122935
NIH Supplement R15MH122935-01S1
COBRE P20GM103650
R01MH131615

Title: Working memory and Abstract cognitive task sequences rely on separable resources

Authors: *M. E. BERRYHILL[1], J. E. SHIRES[1], J. PABLO[1], T. M. DESROCHERS[2];
[1]Psychology, Univ. of Nevada, Reno, NV; [2]Dept. of Neurosci., Brown Univ., Providence, RI

Abstract: Daily life involves accomplishing overarching abstract goals (e.g., have a successful day) with subordinate tasks (e.g., shower, get dressed). Typical visual working memory (WM) tasks involve retention and manipulation of concrete elements within the stimulus array - such as reporting the orientation of a line segment, and tap lateral frontal-parietal activity. A newer paradigm, termed abstract cognitive task sequences (ACTS), requires participants to retain a higher-order series of instructions applied to stimuli as they appear. Thus the WM component is for the instructions, rather than the stimuli. For example, in ACTS participants might report the {color, color, shape, shape} of colored shapes. Importantly, imaging studies reveal that ACTS relies on the rostral lateral prefrontal cortex and shows ramping activity as the participant steps through the sequence. This study was designed to capture the hierarchical nature of WM in maintaining abstract goals and concrete representations. If these are maintained in separate neural representations, increasing demands would not impair performance. Whereas maintaining both in the same neural representation would be capped by WM capacity limits. To test this, we created an ACTS-WM paradigm which doubled WM demands: participants maintained instructions and stimuli. Participants encoded a four-item instruction sequence and applied them to sequentially presented visual stimuli, only reporting answers after stimuli were presented. We predicted that the increased cognitive load would negatively affect performance in the ACTS-WM task. 135 undergraduates performed the ACTS and ACTS-WM tasks online. There were few differences in performance between the two tasks. Both tasks replicated previous reports showing a significant switch-cost within a sequence ($ps < 0.05$), a sequence initiation cost in reaction time ($ps < 0.001$), and reaction time benefit for the last item in the sequence ($ps < 0.001$). Subtle differences emerged between tasks in terms of which ACTS/ACTS-WM sequences elicited superior performance. In summary, interleaving abstract WM goals with concrete stimulus-specific WM responses does not impose a WM cost, possibly by relying on separable frontal correlates.

Disclosures: M.E. Berryhill: None. **J.E. Shires:** None. **J. Pablo:** None. **T.M. Desrochers:** None.

Poster

PSTR169. Distributed Mechanisms of Working Memory and Decision-Making I

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR169.12/PP3

Topic: H.05. Working Memory

Title: Subcortical Structures insights in verbal Working memory stages using EEG

Authors: *S. RANJAN, L. KUMAR;
Electrical Engin., Indian Inst. of Technol. Delhi, Delhi, India

Abstract: Studying verbal working memory (vWM) stages (encoding, maintenance (recall), and retrieval) underpin an individual's cognitive development. The subcortical regions, majorly hippocampus, amygdala, and thalamus, are pivotal in memory function. There is dearth of work exploring temporal dynamics of these structures' involvement in vWM and EEG modality have edge over others on it.
The study employed verbal working paradigms that allow within-subject source activation comparison during vWM stages. In each trial, initially, five targets word and after a delay of 8s, subjects were shown a series of nine probe words. The analysis was performed on 150 trials, recorded from fifteen subjects (10 each) using a 31-channel EEG system. To capture the active source distribution dynamics in the targeted regions, sLORETA-based brain source localization was performed on EEG data using Brainstorm Toolbox. The cortical and deep structure based mixed-head model was used for each subject. "ICBM152 MRI template" and "aseg atlas", the default subject anatomy of Brainstorm is employed to compute the head model for cortical and subcortical structures, respectively, using OpenMEEG BEM. The three-layer boundary element method was used to compute the lead field matrix, with source orientations constrained for the cortical and hippocampus while unconstrained for the amygdala and thalamus. Later, sLORETA was performed to find the spatiotemporal dynamics of the sources. The spatiotemporal dynamics of each targeted region were used to compute the signal change (SC) profile for the vWM stages measured against baseline (rest). Each region's SC profile is considered to understand its involvement, significance, and lateralization in the vWM stages. The significance of the results is estimated using the ANOVA test for $p < 0.05$. The mean amplitude activation for vWM stages is higher in the hippocampus region ($p = 0.0059$), followed by the thalamus ($p = 0.016$) and amygdala ($p = 0.12$), suggesting the hippocampus-thalamic pathology interactions during memory processes. The left and right hippocampus showed higher mean activation amplitudes during encoding and retrieval than maintenance. Additionally, the left hippocampus activation is greater than the right, supporting the idea of the left brain being more verbal (Broca's and Wernicke's area) than the right brain. This is in line with the findings in the study [1] done using fMRI. In particular, the hippocampus is an active and suitable node for the vWM stages.
[1] Karlsgodt, Katherine H., et al. "Hippocampal activations during encoding and retrieval in a verbal working memory paradigm." *Neuroimage* 25.4 (2005): 1224-1231.

Disclosures: S. Ranjan: None. **L. Kumar:** None.

Poster

PSTR169. Distributed Mechanisms of Working Memory and Decision-Making I

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR169.13/PP4

Topic: H.05. Working Memory

Support: USAFOSR FA9550-22-1-0532

Title: A dendrite-based model of the storage of novel, graded-amplitude inputs in working memory

Authors: *J. XU, M. GOLDMAN, S. LUCK, D. COX;
Univ. of California, Davis, Davis, CA

Abstract: Working memory is a fundamental component of many cognitive functions, including reasoning, recall, and decision-making. A key characteristic of working memory is its flexibility, as it allows for the active retention of a novel input without relying on pre-existing learned attractors. Previous models of the storage of novel inputs in working memory have primarily focused on binary memories in which inputs or features are characterized as either present or not present. Here we construct a network model capable of storing a novel, graded-amplitude input in working memory. The model consists of a randomly connected network of neurons, each with an integrate-and-fire soma connected to multiple dendrites. Building upon previous work showing how networks of neurons with NMDA-mediated plateau potentials can store novel binary inputs, each dendrite is endowed with robust voltage bistability mediated by NMDA and GABA-B conductances.
We show that the network can maintain a graded novel input over a large range of input patterns. To understand this result analytically, we map the spiking model onto a rate-based model with all-to-all connectivity. When the pattern of external inputs to the network obeys a linearly decreasing distribution, we show analytically that the network not only maintains firing in the set of neurons that were stimulated, but also their precise intensities. In other words, the network perfectly stores the novel input pattern (up to discretization by the number of independent dendritic compartments). For an input pattern not obeying this ideal distribution, we show that the relative input intensities can still be maintained with a pattern of errors related to the uniform structure of the connectivity matrix and the deviations of the input pattern from the linearly decreasing pattern of intensities. Altogether, this work provides a biophysical network mechanism for encoding a graded, novel input in working memory.

Disclosures: **J. Xu:** None. **M. Goldman:** None. **S. Luck:** None. **D. Cox:** None.

Poster

PSTR169. Distributed Mechanisms of Working Memory and Decision-Making I

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR169.14/PP5

Topic: H.05. Working Memory

Support: NIH R01 EY028746

Title: Fmri neurofeedback biases the maintenance of irrelevant items in working memory

Authors: *D. WHITMER[1], E. S. LORENC[2], Z. BRETTON[3], E. OBLAK[4], J. A. LEWIS-PEACOCK[5];
[1]Univ. of Texas at Austin, Austin, TX; [2]Brown Univ., Providence, RI; [3]Inst. For Neurosci., Univ. Of Texas at Austin, Austin, TX; [4]RIKEN Inst., Wako, Saitama, Japan; [5]Psychology, Univ. of Texas, Austin, Austin, TX

Abstract: The deactivation of irrelevant representations in working memory is beneficial for goal-directed behavior. In retro-cueing tasks, neural representations of un-cued items typically attenuate while cued item representations remain activated. Our study investigates whether neurofeedback can be used to bias working memory representations contrary to primary task demands and thereby influence performance. *Methods*: In a multi-day fMRI study (N = 16), we tested whether fMRI neurofeedback based on the activation of un-cued items in working memory could bias their maintenance. Multi-voxel patterns in the ventral temporal cortex during viewing of face, scene, and object images were used to build subject-specific image category classifiers that were applied during subsequent neurofeedback sessions. Neurofeedback was based on delay-period activity and provided at the end of each trial in a working memory retrocue task that included one face and one scene image. In one condition ('isolation'), the neurofeedback reinforced a low level of delay-period activation for the un-cued item, with the intention of decreasing its activation and isolating it from the cued item. In a second condition ('competition'), the neurofeedback reinforced a higher level of activation for the un-cued item, with the intention of increasing its activation and potentially inducing competition with the cued item. We predicted that this neurofeedback-induced competition would cause long-term forgetting of the un-cued item. The experiment concluded with a surprise recognition memory test for the un-cued items. *Results*: The neurofeedback procedure was successful - the amount of un-cued evidence was higher for 'competition' trials than for 'isolation' trials ($p < 0.05$) whereas cued evidence was unchanged. As expected, the level of classifier evidence for the cued items was predictive of working memory performance for those items. Contrary to our hypothesis, however, modulation of the un-cued items in working memory did not differentially affect their long-term memory outcomes. *Conclusions*: This study demonstrated that fMRI neurofeedback can covertly modulate task-irrelevant working memory representations. While we did not find any consequences for subsequent memory, further work will investigate whether this neuromodulation can meaningfully influence behavior.

Disclosures: D. Whitmer: None. **E.S. Lorenc:** None. **Z. Bretton:** None. **E. Oblak:** None. **J.A. Lewis-Peacock:** None.

Poster

PSTR169. Distributed Mechanisms of Working Memory and Decision-Making I

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR169.15/PP6

Topic: H.05. Working Memory

Support: Swiss National Science Foundation Grant PCEFP1_181141
Jacobs Foundation Research Fellowship 2021-1417-00
NIH Grant 5F32MH115597
NIH Grant K99 MH128893

Title: Decoding children's working memory contents with EEG

Authors: *N. TUROMAN[1], P. A. FIAVE[1], C. ZAHND[1], M. T. DEBETTENCOURT[2], E. VERGAUWE[1];
[1]Univ. de Genève, Geneva, Switzerland; [2]Univ. of Chicago, Univ. of Chicago, Chicago, IL

Abstract: Working memory (WM) is an important predictor of cognitive development and educational outcomes. WM performance improves with age, and such improvement has been linked to the developmental onset of using specific maintenance strategies spontaneously. However, accurately measuring this onset has been difficult, as typically-used behavioural methods rely on task manipulations which can bias the detection and operation of the mechanisms of interest. We thus aimed to develop and verify a more direct measure of children's WM maintenance that is free of additional task manipulations and that allows children to maintain information spontaneously. Specifically, electroencephalography (EEG) data was collected from 20 children (7 female, age range: 7-12 years, mean age 9 years and 7 months) while they played a simple computerised WM game without any instructions on how memoranda should be maintained. Multivariate Pattern Analysis (MVPA) was used to examine if 3 categories of maintained information (visual, spatial, and verbal) could be distinguished from EEG signals when memoranda were observed (Sensory period) and maintained (Delay period). Average classification accuracy was significantly above chance (33%), both during the Sensory (58%) and Delay periods (44%). Further, classification patterns generalised over time, such that representations detected during the Sensory period could be traced for a portion of the Delay period. These results held even with half as much data, were consistent across individuals, and were not driven by older participants with presumably high WM performance. Thus, in a first study of its kind, we showed that children's brains represent different category information, and that the representations that were formed when memoranda were observed persisted in WM maintenance. Further, we show that combining a simple child-friendly task with MVPA of EEG data can detect and track differences in observed and maintained information, in a robust and consistent manner. This exploratory project highlights the sensitivity of MVPA for investigating WM content, and opens the door to further investigations into the specific mechanisms used to maintain said content in children.

Disclosures: N. Turoman: None. **P.A. Fiave:** None. **C. Zahnd:** None. **M.T. deBettencourt:** None. **E. Vergauwe:** None.

Poster

PSTR169. Distributed Mechanisms of Working Memory and Decision-Making I

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR169.16/PP7

Topic: H.05. Working Memory

Support: R56 MH125642
R01 MH129042
T32 MH106454

Title: Suppressing thoughts from working memory can produce enduring changes to their memory trace

Authors: *Z. BRETTON[1], H. KIM[2], M. T. BANICH[2], J. A. LEWIS-PEACOCK[3];
[1]The Inst. for Neurosci., Univ. Of Texas at Austin, Austin, TX; [2]Univ. of Colorado Boulder, Univ. of Colorado Boulder, Boulder, CO; [3]Univ. of Texas at Austin, Univ. of Texas, Austin, Austin, TX

Abstract: Our work explores the intentional removal of thoughts from working memory and its potential long-term consequences on memory. Replacing thoughts and suppressing thoughts are each associated with distinct neural processes and consequences. Replacing one thought with another quickly removes the unwanted thought out of the focus of attention, but this alone does not reduce its accessibility in working memory. Suppressing a thought requires sustained focal attention on that information, which can reduce its availability, thus freeing up capacity in working memory. Beyond the scope of working memory, the lasting effects of these removal operations are yet uncertain. Drawing upon the sensory recruitment model of working memory, we hypothesized that deliberately removing representations of visual stimuli from working memory would modify their sensory representations retained in the visual cortex, which may result in subsequent forgetting. To investigate this, we conducted an fMRI study (N = 25) where participants performed a working memory removal task using images of natural and manmade scenes. Following the task, a surprise recognition test assessed participants' long-term memory of the examined items. Using multivariate pattern classification and representational similarity analysis of the fMRI data, we assessed the impact of removal operations on the neural representation of items and linked the neural evidence to subsequent memory. Our findings revealed that the engagement of suppression led to stronger activation of a given item in working memory and also to its subsequent forgetting. Furthermore, successfully suppressing an item in working memory led to enduring alterations in its item-level features but not its category-level features, contributing to its subsequent forgetting. These results suggest that actively removing an object from working memory through suppression may weaken its representation in long-term memory by diminishing item-specific features, potentially leading to the item's eventual forgetting.

Disclosures: Z. Bretton: None. **H. Kim:** None. **M.T. Banich:** None. **J.A. Lewis-Peacock:** None.

Poster

PSTR169. Distributed Mechanisms of Working Memory and Decision-Making I

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR169.17/PP8

Topic: H.05. Working Memory

Support: NIH R01 MH123679

Title: Genuine beta bursts in human working memory: controlling for the influence of lower-frequency rhythms

Authors: *J. RODRIGUEZ-LARIOS[1], S. HAEGENS[2];
[2]Dept. of Psychiatry, Div. of Systems Neurosci., [1]Columbia Univ., New York, NY

Abstract: Human working memory is associated with significant modulations in oscillatory brain activity. However, the functional role of brain rhythms at different frequencies is still debated. Modulations in the beta frequency range (15-40 Hz) are especially difficult to interpret because they could be artifactually produced by (more prominent) oscillations in lower frequencies that show non-sinusoidal properties. In this study, we investigate beta oscillations during working memory while controlling for the possible influence of lower frequency rhythms. We collected electroencephalography (EEG) data in 31 participants who performed a spatial working-memory task with two levels of cognitive load. In order to rule out the possibility that observed beta activity was affected by non-sinusoidalities of lower frequency rhythms, we developed an algorithm that detects transient beta oscillations that do not coincide with more prominent lower frequency rhythms in time and space. Using this algorithm, we show that the amplitude and duration of beta bursts decrease with memory load and during memory manipulation, while their peak frequency and rate increase. In addition, interindividual differences in performance were significantly associated with beta burst rates. Together, our results show that beta rhythms are functionally modulated during working memory and that these changes cannot be attributed to lower frequency rhythms with non-sinusoidal properties.

Disclosures: J. Rodriguez-Larios: None. **S. Haegens:** None.

Poster

PSTR169. Distributed Mechanisms of Working Memory and Decision-Making I

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR169.18/PP9

Topic: H.05. Working Memory

Support: NIH Grant F32MH124268

Title: Differential causal roles of parietal cortex, frontal cortex and cerebellum in spatial working memory

Authors: *J. A. BRISSENDEN, T. G. LEE;
Univ. of Michigan, Ann Arbor, MI

Abstract: Working memory (WM) enables the short-term maintenance of mental representations in support of ongoing cognitive operations. Despite extensive research focused on identifying the specific brain structures and mechanisms involved in WM, much of the prior literature has relied on correlational methods to explore the neural substrates of WM. We sought to investigate the causal role of brain areas spanning the cerebral cortex and cerebellum in WM using a combination of functional magnetic resonance imaging (fMRI) and transcranial magnetic stimulation. Across multiple sessions, we applied continuous theta-burst stimulation (cTBS) to perturb activity in intraparietal sulcus, frontal eye fields, cerebellar lobule VIIb, and a somatosensory control site immediately prior to the performance of a continuous report spatial working memory task both inside and outside of the fMRI scanner. A baseline fMRI session collected structural MRI and functional scans for resting-state and population receptive field mapping. We examined the effect of cTBS on parameters of a variable precision mixture model fit to the recall error distribution for each session. We also investigated how the disruption of each area affected the probabilistic decoding of remembered location from BOLD activity across the brain. Cerebellar and frontal cTBS resulted in reduced recall precision relative to control stimulation, while intraparietal sulcus stimulation increased precision variability. We also demonstrated that stimulation of our regions of interest decreased decoding accuracy and increased decoding uncertainty within downstream visual cortical areas relative to the control site. Further analysis attributed the impact of cTBS on decoding to an increase in the variance of spatially tuned responses to repeated presentations of the same stimulus. These results provide evidence for a causal contribution to working memory maintenance by a distributed network of areas spanning both cerebral cortex and cerebellum.

Disclosures: J.A. Brissenden: None. **T.G. Lee:** None.

Poster

PSTR169. Distributed Mechanisms of Working Memory and Decision-Making I

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR169.19/PP10

Topic: H.05. Working Memory

Support: R01NS116589

Title: Multiplexing working memory and timing: Sequential activity in CA1 during a novel differential delayed nonmatch-to-sample (dDNMS) task.

Authors: *P. GOLSHANI[1], D. BUONOMANO[2], J. TAXIDIS[3], C. DORIAN[4];
[1]UCLA Dept. of Neurol., Los Angeles, CA; [2]UCLA, Los Angeles, CA; [3]Neurosciences and Mental Hlth., The Hosp. For Sick Children, Toronto, ON, Canada; [4]Univ. of California, Los Angeles Interdepartmental Ph.D. Program In Neurosci., Los Angeles, CA

Abstract: Working memory (WM) is the system for temporary storage and manipulation of information to guide behavior. Implicit timing refers to the ability of animals to learn the

temporal structure of external events, even though timing may be irrelevant to the task itself. Recent studies, for example, indicate that humans implicitly learn the timing of delays during WM tasks (Zhou et al., 2023). Even though WM and timing may be highly intertwined, these two fields of studies have largely been explored separately because they are often seen as unique cognitive functions with different underlying neural mechanisms. Since WM requires the transient storage of retrospective information, while timing transiently generates prospective information, we asked if WM and timing, in some cases, may rely on shared neural mechanisms. To answer this question, we developed a novel differential-delayed nonmatch-to-sample (dDNMS) task, in which the identity of the first odor stimulus predicts the delay duration (e.g., AB and AA trials have 2.5 second delays; BA and BB trials have 5 second delays). While the cued-differential delays are irrelevant to the WM task itself, it allows us to probe anticipation and implicit timing. Behavioral data suggests that mice learn the implicit timing component of the task because there is a performance deficit (n=12, p=0.021) when cue-delays are switched, thus violating their temporal expectation. To investigate whether previously observed sequential neural activity in the hippocampus (Taxidis et al., 2020) may encode both WM and timing, we have conducted 2-photon calcium imaging of CA1 pyramidal neurons expressing GCaMP7f, while mice performed the dDNMS task. Strong odor and time information is encoded at the individual cell and population level activity with 20% of neurons encoding odor during odor presentation and 14% encoding time - as defined by cells having a significant firing field during the delay period. Most interestingly, the velocities of neural trajectories were different during the first 2.5 seconds of short vs long delays. An acceleration occurs preceding the onset of the 2nd odor, suggesting anticipation and that CA1 sequential activity may be encoding both WM (odor identity) and expected onset of the second odor (implicit timing). Ongoing experiments will evaluate how these dynamics emerge over learning, and how CA1 maintains working memory odor information when implicit timing expectations are violated.

Disclosures: **P. Golshani:** None. **D. Buonomano:** None. **J. Taxidis:** None. **C. Dorian:** None.

Poster

PSTR169. Distributed Mechanisms of Working Memory and Decision-Making I

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR169.20/PP11

Topic: H.06. Social Cognition

Support: AMED (JP21wm0525001)
JSPS-KAKENHI (22H04931)
JSPS-KAKENHI (19H05467)
JST FOREST Program (JPMJFR224B)

Title: Distinct roles of two frontal areas in action prediction

Authors: *T. NINOMIYA[1,2], M. ISODA[1,2];
[1]Natl. Inst. for Physiological Sci., Okazaki, Japan; [2]Grad. Inst. for Advanced Studies, SOKENDAI, Okazaki, Japan

Abstract: Monitoring others' actions is crucial for successful social exchanges. Predicting others' actions is a key aspect of such social monitoring functions, but its neural basis is yet to be determined. In the present study, we sought to address how the ventral premotor cortex (PMv) and the medial prefrontal cortex (MPFC), frontal core nodes in social brain networks, are involved in the prediction of others' actions. To this end, we trained macaque monkeys to perform a role-reversal choice task. In this task, a monkey undergoing neural recordings (designated as M1) faced a partner monkey (designated as M2) indirectly through a video device. This device allowed us to manipulate visual information of M2's actions available for M1 while ensuring real-time communication between the subjects. M1 and M2 alternated the roles of "actor" and "observer" every two trials. In each trial, one, two, or three target buttons were illuminated for the actor to make a choice, and both subjects were rewarded with a drop of water when the actor chose the correct target. The number of the targets and the location of the correct target remained the same for a block of 6 trials. In approximately 15% of M2's actor trials, the reaching movement was entirely occluded from M1's sight using the video device (occluded condition). We examined if the PMv and MPFC were involved in two aspects of action prediction, i.e., prediction of upcoming actions and prediction of ongoing actions, by quantifying neuronal activities before and during the time at which M2 would have made a choice. Activities of isolated neurons were initially classified into three types (self, mirror, or partner type) based on their specificity to agents at the time of choice. Neuronal activities were then compared between the full vision condition and the occluded condition. We found that choice-related activities were mostly comparable between the occluded condition and the full vision condition for mirror-type and partner-type neurons in both areas. On the other hand, a subset of partner-type neurons in the MPFC, but not in the PMv, modulated their activities immediately after the onset of the occlusion. These early-onset MPFC partner-type neurons exhibited either larger or lower activities when M1 was more difficult to predict M2' choice (i.e., in the first trial of each block with larger number of illuminated targets). These results suggest that the PMv and MPFC are primarily involved in predicting ongoing and upcoming action of others, respectively.

Disclosures: T. Ninomiya: None. **M. Isoda:** None.

Poster

PSTR170. Episodic and Episodic-Like Memory-Neurophysiology and Models

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR170.01/PP12

Topic: H.07. Long-Term Memory

Support: NIH grant 5R01MH078064
NIH grant 5R01MH108837
FWF J 4271

Title: Encoding Temporal Traces within Hippocampal-Cortical Circuits

Authors: *A. CICVARIC[1], T. BASSETT[1], Z. PETROVIC[1], L. REN[2], V. JOVASEVIC[2], A. L. GUEDEA[2], K. PARKER[1], V. GRAYSON[2], N. YAMAWAKI[3], G. M. SHEPHERD[2], J. M. RADULOVIC[1,2,3];
[1]Albert Einstein Col. of Med., Bronx, NY; [2]Northwestern Univ., Chicago, IL; [3]Univ. of Aarhus, Aarhus, Denmark

Abstract: Episodic memories are integrated representations of past experiences of events, their temporal relationships, as well as places where they occurred. Here, we used chemogenetic and fiber photometry approaches in combination with trace fear conditioning paradigm (TFC) to study the mechanisms contributing to the encoding of temporal traces, short time intervals that separate two sequences of stimuli. Excitatory neurotransmission in the dorsal hippocampus (DH) together with several cortical areas, including retrosplenial cortex (RSC), has been shown to play a key role in TFC. We previously dissected the DH→RSC circuit and found two molecularly distinct projections that express either VGLUT1 or VGLUT2. Here, we showed that both types of projections were necessary the formation of tone-trace-shock associations, whereas the processing of the trace itself was dependent solely on the VGLUT2-containing projections. Furthermore, fiber photometry recordings of the DH→RSC VGLUT2-containing projections showed distinct activity patterns during memory encoding and retrieval. We are currently investigating the postsynaptic components of the DH→RSC circuit and how the differences in the activity of these projections are contributing to the activity of RSC neurons during TFC.

Disclosures: A. Cicvaric: None. **T. Bassett:** None. **Z. Petrovic:** None. **L. Ren:** None. **V. Jovasevic:** None. **A.L. Guedea:** None. **K. Parker:** None. **V. Grayson:** None. **N. Yamawaki:** None. **G.M. Shepherd:** None. **J.M. Radulovic:** None.

Poster

PSTR170. Episodic and Episodic-Like Memory-Neurophysiology and Models

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR170.02/PP13

Topic: H.07. Long-Term Memory

Support: NIMH MH108837
NIMH MH078064

Title: Tgf-beta signaling stabilizes memory circuits during adolescence

Authors: *H. ZHANG[1], Z. PETROVIC[1], K. PARKER[1], A. CARBONCINO[1], E. WOOD[1], V. JOVASEVIC[2], A. L. GUEDEA[2], P. YI[2], J. M. RADULOVIC[1,2];
[1]Dominick P. Purpura Dept. of Neurosci., Albert Einstein Col. of Med., Bronx, NY; [2]Dept. of Psychiatry and Behavioral Sci., Northwestern Univ., Chicago, IL

Abstract: The persistence of episodic memory requires interactions between the hippocampus and selected cortical areas. It is believed that hippocampal-cortical circuits are formed during the postnatal developmental period and stabilized during adolescence. The brain undergoes important changes during the adolescent period leading to enhanced stress sensitivity and enhanced emotional responses. It is not known, however, whether and in what way these changes might affect the memory circuits representing stressful experiences. To address these questions, we examined the neurobiological mechanisms of stress-related memories before, during, and post adolescence. We used the contextual and trace fear conditioning paradigms and analyzed the maturation of the projections from the dorsal hippocampus (DH) to retrosplenial cortex (RSC) as well as various parameters of neuron-specific, memory-related cellular signaling. We found that the DH-RSC circuit undergoes significant developmental changes in the investigated time period that were paralleled with fluctuations in performance. Cellular The observed effects could be stabilized, at least in part, through TGF-beta signaling. manipulations within this circuit suggested that the performance variability was most likely caused by changes in accessibility of stress-related memories to retrieval.

Disclosures: H. Zhang: None. **Z. Petrovic:** None. **K. Parker:** None. **A. Carboncino:** None. **E. Wood:** None. **V. Jovasevic:** None. **A.L. Guedea:** None. **P. Yi:** None. **J.M. Radulovic:** None.

Poster

PSTR170. Episodic and Episodic-Like Memory-Neurophysiology and Models

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR170.03/PP14

Topic: H.07. Long-Term Memory

Support: NIH Grant 1R01MH108837
NIH Grant 1R01MH078064

Title: Inflammatory Mediators of Memory Formation

Authors: *E. M. WOOD[1], Z. PETROVIC[1], A. CARBONCINO[1], K. PARKER[1], H. ZHANG[1], A. CICVARIC[1], J. M. RADULOVIC[1,2,3];
[1]Albert Einstein Col. of Med., Bronx, NY; [2]Aarhus Univ., Aarhus, Denmark; [3]Northwestern Univ., Chicago, IL

Abstract: Neuroinflammation, characterized by increased microglial and astrocytic activation and increased cytokine signaling, is a hallmark of various neurological diseases, often adversely affecting learning and memory. However, numerous studies also demonstrate that inflammatory signaling is necessary for memory formation. The nuclear factor kappa B (NFκB) family, transcription factors classically known as master immune regulators, has previously been shown to regulate gene expression in neurons for structural plasticity and long-term memory. How NFκB balances or combines its functional roles in immune regulation and memory formation is unknown. We demonstrate that in the mouse hippocampus, neuronal, but not astrocytic

knockdown of RelA, the most abundant NFκB family member, impairs contextual memory formation. Additionally, we show that neuronal knockdown of a RelA downstream target, interferon-α/β receptor 1 (IFNAR), leads to deficits in contextual fear memory. We also demonstrate that IFNAR knockdown results in increased size and nuclear accumulation of learning induced RelA. Given that NFκB/RelA induces interferon transcription, our results suggest that interferon signaling provides negative feedback to downregulate RelA following learning. Thus, our findings show that contrary to the negative effects of astrocytic/glial inflammatory signaling on learning and memory, discrete components of neuronal inflammatory pathways support learning and memory and have a pro-cognitive effect.

Disclosures: E.M. Wood: None. **Z. Petrovic:** None. **A. carboncino:** None. **K. Parker:** None. **H. Zhang:** None. **A. Cicvaric:** None. **J.M. Radulovic:** None.

Poster

PSTR170. Episodic and Episodic-Like Memory-Neurophysiology and Models

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR170.04/PP15

Topic: H.07. Long-Term Memory

Support: NIH Grant 5R01MH078064
NIH Grant 5R01MH108837

Title: Social Memory Integration in the Medial Temporal Lobe

Authors: *T. E. BASSETT[1], A. CICVARIC[1], Z. PETROVIC[1], A. CARBONCINO[1], J. M. RADULOVIC[1,2,3,4];
[1]Dominick P. Purpura Dept. of Neurosci., [2]Dept. of Psychiatry and Behavioral Sci., Albert Einstein Col. of Med., The Bronx, NY; [3]Dept. of Biomedicine, Aarhus Univ., Aarhus, Denmark; [4]Dept. of Psychiatry and Behavioral Sci., Northwestern Univ., Chicago, IL

Abstract: Abnormal processing of social interactions, including the formation of social memories, has a lasting negative impact on mental health and it is found many psychiatric illnesses, ranging from neurodevelopmental disorders to social anxiety disorder and depression. Therefore, uncovering the underlying neurobiological mechanisms behind the processing of social information is critical to understanding the development of these harmful and persistent affective states. The Hippocampal-Entorhinal Cortex complex plays a well-known role in the formation and integration of memories, particularly for the relational, temporal, and contextual components of a memory representation. More recently, the complex's known role was expanded to include social memory, with the lateral entorhinal cortex (EC) projection into the dCA2 being proven necessary for social recognition, a key component of social memory. To test whether CA2-EC interactions are also required for the formation and recall of long-term social memories, we are using the Social Fear Conditioning (SFC) paradigm in conjunction with an array of circuit tracing and manipulations. Our findings demonstrate that SFC induces robust freezing to social

stimuli that can be readily differentiated from the contextual and cue-related freezing acquired in the same episode. Ongoing experiments are interrogating the neurobiological mechanisms underlying the encoding of these different memory components.

Disclosures: **T.E. Bassett:** None. **A. Cicvaric:** None. **Z. Petrovic:** None. **A. carboncino:** None. **J.M. Radulovic:** None.

Poster

PSTR170. Episodic and Episodic-Like Memory-Neurophysiology and Models

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR170.05/PP16

Topic: H.07. Long-Term Memory

Support: UCLA Dean Life Recruitment and Retention Grant 404040-CL-89313

Title: Overlapping memories are distorted under arousal and anxiety

Authors: *E. MORROW, D. V. CLEWETT;
Psychology, UCLA, Los Angeles, CA

Abstract: In our daily lives, we often encounter highly similar information. One way to prevent these memories from clashing with each other is memory repulsion, a learning-dependent process by which subtle differences between memories become exaggerated to reduce interference. Although these repulsion effects have been observed under neutral conditions, it is less clear how such distortions are affected by emotional arousal - when resolving interference is perhaps most needed. Here, we adapted an existing behavioral paradigm in young adults to examine if arousal influences memory repulsion between similar events, and whether such distortions relate to lower memory interference. In this task, participants viewed different face-object pairs. There were two versions of each object (a *target* and *competitor*) that differed slightly in color to induce interference. For half of the participants, pairmate colors will be highly similar (24° apart), whereas for the other half of participants, the pairmate colors will be less similar (72° apart). Additionally, the pairmates (*competitors*) were either preceded by a neutral tone or aversive white noise burst to manipulate arousal. Distortions in color memory were tested for the remaining neutral pairmates (*targets*) on a circular color wheel, while memory interference was assessed using face-object association memory tests. Preliminary results showed a trending effect of arousal on color memory that differed by perceptual similarity ($F(1,53) = 3.26$, $p = .077$). Contrary to our expectations, highly similar pairs were attracted, or rated closer to each other on the color wheel when associated with arousal ($t(31) = -1.96$, $p = .059$), while dissimilar pairs were not ($t(22) = 0.70$, $p = .49$). We also replicated previous findings showing that as attraction intensifies overall, memory interference increases ($\rho = -0.40$, $p = .003$). Interestingly, individuals with higher trait anxiety showed a bias towards arousal-related attraction ($\rho = -0.29$, $p = .03$), suggesting that psychiatric symptoms may blur the overlap between related negative and neutral memories. Together, our initial findings suggest that

arousal may further obscure the differences between highly similar memories. Additional pupillometry analyses will examine if arousal mediates the relationship between memory distortions and memory interference.

Disclosures: E. Morrow: None. **D.V. Clewett:** None.

Poster

PSTR170. Episodic and Episodic-Like Memory-Neurophysiology and Models

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR170.06/Web Only

Topic: H.07. Long-Term Memory

Support: UNAM DGAPA PAPIIT IG300121

Title: Alcohol consumption effects on episodic memory across the adult lifespan

Authors: *S. CANSINO[1], F. TORRES-TREJO[1], C. ESTRADA-MANILLA[1], S. RUIZ-VELASCO[2];
[1]Lab. of NeuroCognition, [2]Applied Mathematics and Systems Res. Inst., Univ. Nac Autónoma de México, Mexico City, Mexico

Abstract: The aim of the present study was to establish whether moderate alcohol intake benefits episodic memory accuracy and speed in a lifespan sample after classifying individuals as lifetime nondrinkers and drinkers and controlling for several demographic and biological variables. A sample of 1,557 healthy adults between 21 and 80 years of age participated in the study. Alcohol consumption was assessed through a lifestyle questionnaire created for the study. Episodic memory performance was measured through a computerized task that allowed us to reliably measure recollection, the most vulnerable process within episodic memory as age advances. Recollection accuracy was superior in drinkers than in nondrinkers. Hierarchical regression models demonstrated that the total alcohol intake and the amount of alcohol and wine intake were associated with higher recollection. Beer drinkers showed more accurate and faster responses in recollection and recognition than spirit drinkers and nondrinkers. The major benefit on recollection and recognition accuracy was reached with 150 grams of alcohol per week for women and men, which corresponded to 11 drinks per week. Although alcohol consumption has been considered a health risk, at least for episodic memory, we found that moderate alcohol intake has more beneficial effects across the entire adult lifespan than alcohol abstinence.

Disclosures: S. Cansino: None. **F. Torres-Trejo:** None. **C. Estrada-Manilla:** None. **S. Ruiz-Velasco:** None.

Poster

PSTR170. Episodic and Episodic-Like Memory-Neurophysiology and Models

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR170.07/PP17

Topic: H.07. Long-Term Memory

Support: National Institutes of Health Research Project Grant (R01EY034436)
Zuckerman Institute Seed Grant for MR Studies (CU-ZI-MR-S-0016)

Title: Prospective and retrospective representations of temporal structure across hippocampal and visual regions

Authors: *H. TARDER-STOLL, C. BALDASSANO, M. ALY;
Columbia Univ., New York, NY

Abstract: Memories for sequences of events allow us to anticipate future experiences at multiple timescales. Past studies of multistep anticipation has shown a prospective hierarchy, with anticipation of progressively further events represented in progressively more anterior brain regions. However, it remains unknown whether such a processing hierarchy extends bidirectionally into the past and the future during anticipation of temporal structure. Participants (N = 32) learned sequences of environments in immersive virtual reality and then, during fMRI, anticipated upcoming environments multiple steps into the future in the learned sequence. Using multivariate fMRI analyses, we show evidence for both prospective and retrospective representations of sequences during a multistep anticipation task. Such representations were (1) graded, with successively less evidence for environments further away in the sequence, (2) organized hierarchically, with shorter timescales in visual cortex and longer timescales in hippocampus, and (3) related to participants' behavioural ability to make anticipatory judgements. We further found preliminary evidence for hierarchical representations of bidirectional temporal structure within ventral visual regions along a posterior to anterior axis, such that anterior aspects of regions had successively more graded representations of temporal structure than posterior aspects of regions. Together, our results shed light on how complex, extended sequences are represented across the brain and used to guide adaptive and flexible behaviour.

Disclosures: H. Tarder-Stoll: None. **C. Baldassano:** None. **M. Aly:** None.

Poster

PSTR170. Episodic and Episodic-Like Memory-Neurophysiology and Models

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR170.08/PP18

Topic: H.07. Long-Term Memory

Title: Is transcutaneous vagus nerve stimulation an effective modulator of the LC-NE system? Testing human enzymatic, electrophysiological, and behavioral outcome measures

Authors: *H.-V. NGO[1], I. HEYDE[2], H. OSTER[2], J. OBLESER[2];
[1]Univ. of Lübeck, Lübeck, Germany; [2]Univ. of Luebeck, Luebeck, Germany

Abstract: The noradrenaline (NE) system emerging from the locus coeruleus (LC) is thought to regulate arousal or cognitive processes and aberrances of the LC-NE system underlie pathological conditions like depression or epilepsy. Given this importance, transcutaneous vagal nerve stimulation (tVNS) has continuously attracted attention as a non-invasive technique to target the LC by stimulating the vagal nerve via the outer ear. Tonic tVNS (a 30s on/off stimulation pattern) has found particularly wide use in clinical settings, with LC-NE modulation reducing epileptic seizures or counteracting symptoms of depression. However, the exact working mechanism are not fully understood. Specifically, little is known about the link between electrophysiological correlates of tVNS and cognitive outcomes. In this explorative study, healthy young subjects (N = 32) performed either an auditory discrimination or a visual associative recognition task under tonic tVNS. Furthermore, electroencephalography and pupillometry were recorded to examine evoked brain responses, pupil dilation and aperiodic brain activity as biomarkers of tVNS and noradrenergic modulation. More importantly, the temporal pattern of the tonic stimulation pattern was utilized to directly contrast tVNS ON vs. tVNS OFF phases within each subject. Contrary to our expectation, tVNS ON and OFF periods showed no effect on auditory or visually evoked potentials and event-related pupil dilation. Similarly, examining the 1/f slope of aperiodic EEG activity revealed no ON/OFF tVNS effect and thus no signature of noradrenergically-driven change in excitation/inhibition balance. This absence in electrophysiological differences is in line with no difference in pitch discrimination ($eta^2 = 0.242$) or recognition performance ($eta^2 = 0.051$) in comparison to a control group exposed to a sham stimulation. Intriguingly, however, saliva levels of alpha-amylase activity - a proxy of noradrenergic activity - obtained via saliva sampling before and after tVNS showed a clear increase after stimulation compared to the sham condition (Cohen's d = 1.04), an effect that appeared specific to the LC-NE system (i.e., no corresponding change in saliva glucocorticoids). Despite the wide use of tonic tVNS, the present exploratory data show a striking dissonance between electrophysiological markers versus alpha-amylase activity as a coarse-grain correlative marker of the sympathetic nervous system. Altogether, these findings highlight the need of reliable and specific biomarkers when dissecting the stimulation parameters and temporal dynamics to understand and utilize the modulative potential of tonic tVNS.

Disclosures: H. Ngo: None. **I. Heyde:** None. **H. Oster:** None. **J. Obleser:** None.

Poster

PSTR170. Episodic and Episodic-Like Memory-Neurophysiology and Models

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR170.09/PP19

Topic: H.07. Long-Term Memory

Support: NSF Grant 1633873

Title: Moderating effects of cortical volume and thickness on retrieval-related scene reinstatement

Authors: *J. M. OLIVIER[1], S. SROKOVA[1], M. D. RUGG[2];
[1]Univ. of Texas at Dallas, Richardson, TX; [2]Univ. of Texas at Dallas, Dallas, TX

Abstract: Cortical reinstatement refers to retrieval-related reactivation of cortical activity patterns that partially overlap with those elicited during the initial encoding of the retrieved episode. Thus, the retrieval of images of visual scenes is associated with reinstatement in canonical scene-selective cortical regions such as the parahippocampal place area (PPA) and the retrosplenial complex (RSC). Additionally, scene reinstatement is robustly weaker in older than young adults. Here, we conducted a series of multiple regression analyses to examine the relationships between scene reinstatement, age, global cortical volume, and global cortical thickness, combining data from two prior experiments in which young (n = 42; 25 female) and older (n = 40; 20 female) adult humans performed one of two source memory tasks while undergoing fMRI. Thickness and volume were significantly lower in the older than the young adult group. In each experiment, participants first studied words presented simultaneously with images of scenes and either faces (experiment 1) or scrambled images (experiment 2). At retrieval, participants in both experiments first judged whether test words were old or new. For each word judged old, they were prompted to signal the image category that the word had been paired with at study (i.e., scene, face, or scrambled). After controlling for experiment and sex, both cortical volume and cortical thickness were significant predictors of the strength of scene reinstatement (operationalized by a univariate metric, the 'reinstatement index') in both the PPA and the RSC. Cortical volume remained a significant predictor of PPA and RSC reinstatement when age group was included in the model, whereas cortical thickness now predicted reinstatement only in the PPA. In a final model which included cortical volume, cortical thickness, age group, and experiment as predictors, cortical volume, but no other variable, predicted a significant fraction of the variance in reinstatement strength in the RSC. By contrast, none of the predictors individually predicted reinstatement in the PPA. Together, these findings suggest that age differences in scene reinstatement in the PPA and RSC are largely mediated by cortical structural variables.

Disclosures: J.M. Olivier: None. **S. Srokova:** None. **M.D. Rugg:** None.

Poster

PSTR170. Episodic and Episodic-Like Memory-Neurophysiology and Models

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR170.10/PP20

Topic: H.07. Long-Term Memory

Support: Deutsche Forschungsgemeinschaft (SA-2146/6-1).

Title: Selective decrease of PER, LEC and spatial hippocampal subnetwork activity during object-in-place remote memory retrieval in memory-impaired mice

Authors: *E. ATUCHA[1], C. FUERST[2], M. SAUVAGE[3];
[1]Leibniz Inst. For Neurobio., Magdeburg, Germany; [2]Leibniz Inst., Magdeburg, Germany;
[3]Functional Architecture of Memory Dpt, Leibniz Inst. for Neurobio., Magdeburg, Germany

Abstract: Main consolidation theories have shown that cortical regions, such as the PFC, are increasingly engaged as memories age. However, little is known about the contribution of cortical regions belonging to the medial temporal lobe (MTL), namely the PER, LEC, MEC, POR, within this frame. We recently showed that the engagement of these cortical areas follows a pattern similar to that of the anterior cingulate cortex for the retrieval of contextual fear and object-location memories up to 1-year old (comparable to 40 years-old in humans based on life expectancy; Lux et al, Elife, 2016, Atucha et al., bioRxiv, 2021,sfn abstract, 2021). Moreover, we reported that the proximal part of CA1 and the distal part of CA3 (both close to CA2) might constitute a functional network within the hippocampus that process preferentially spatial information (Nakamura et al, 2013; Flasbeck et al, 2018; Beer and Vavra et al, 2018). Here we further investigate the contribution of the MTL cortical and PFC areas and that of the spatial hippocampal subnetwork to memory retrieval over half a life span by imaging brain activity in memory-impaired and memory-intact mice performing on an object-location memory task using high resolution Arc immediate-early gene. We found that PER, LEC, MEC, POR and the ACC (but not PrL, nor IL) were increasingly engaged for retrieving recent, early remote and very remote memories in memory -intact mice (1day, 1month and 6 months-old, respectively). In addition, CA1 was persistently activated over this time-window while CA3 was no longer recruited for retrieving the most remote memories. In a striking contrast, a selective decrease in activity could be detected in the PER, LEC and the spatial hippocampal subnetwork subregions (distal CA3 and proximal CA1) in mice showing memory-impairment (starting one month after memory formation) while activity in MEC, POR, ACC and the non-spatial hippocampal subnetwork (proximal CA3 and distal CA1) was comparable to that of memory-intact mice. Similar patterns were observable when memories were even more remote (6 months and 1 year-old memories) but in CA3, which is no longer recruited at this time-point. By investigating memories that naturally lost their precision, we bring here evidence that the LEC, PER, proximal CA1 and distal CA3 play a more important role in retrieving early and very remote object-location memory than their functional counterparts (i.e. the MEC, POR, distal CA1 and proximal CA3).

Disclosures: **E. Atucha:** None. **C. Fuerst:** None. **M. Sauvage:** None.

Poster

PSTR170. Episodic and Episodic-Like Memory-Neurophysiology and Models

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR170.11/PP21

Topic: H.07. Long-Term Memory

Support: R01MH132171
R01MH130374

Title: Jumping Back In Time: A neural network model for episodic memory recollection by pattern completion of a scale-invariant temporal context

Authors: *C. S. HALL[1], Z. G. ESFAHANI[2], P. B. SEDERBERG[1], M. W. HOWARD[2]; [1]Psychology, Univ. of Virginia, Charlottesville, VA; [2]Psychological & Brain Sci., Boston Univ., Boston, MA

Abstract: It is widely believed that episodic memory results from recovery of spatiotemporal context and the hippocampus represents the spatiotemporal context via place cells, time cells, etc. Computational temporal context models have provided a good account of behavioral data in many human episodic recall tasks, but have not yet specified a neural mechanism by which a temporal context could be retrieved. We study a neural network model of episodic memory and its behavior after presenting it with lists of random stimuli. Temporal context is operationalized as populations of time cells triggered by each item in the list. At any moment, the activity of the population codes the recent history of what happened when leading up to that moment. During study of a list, connections between time cells with similar time constants undergo Hebbian plasticity. During retrieval, inhibitory interneurons feed back to the time cells forming a local excitation/global inhibition bump attractor similar to the *Drosophila* head direction circuit. During retrieval of an episodic memory, activity spreads sequentially from one time scale to the next, eventually retrieving the entire context from a particular moment during study. If the time cells tile the log time axis evenly, as recently observed in rodent hippocampus, and if there is appropriate plasticity between time cells and interneurons, then the network is guaranteed to be invariant to changes in the rate at which the list was presented, resulting in a scale-invariant neural network model for episodic memory retrieval.
This neural network model has many properties that map on to experiments in cognitive neuroscience and cognitive psychology. Like temporal context models, the network provides a natural account of recency and contiguity effects. The network is also sensitive to event boundaries, here operationalized as the beginning of a list. States of context just after event boundaries are especially easy to retrieve, providing a principled account of the primacy effect. In addition to providing a bridge between time cells, pattern completion models of the hippocampus, and a broad range of behavioral findings from human episodic memory, this model also makes falsifiable assertions about the organizational connectivity of the hippocampus as well as dynamic patterns of neural activity during retrieval of episodic memory.

Disclosures: C.S. Hall: None. **Z.G. Esfahani:** None. **P.B. Sederberg:** None. **M.W. Howard:** None.

Poster

PSTR170. Episodic and Episodic-Like Memory-Neurophysiology and Models

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR170.12/PP22

Topic: H.07. Long-Term Memory

Title: A model of episodic memory based on similarities to the holographic principle

Authors: *D. KAWAHARA[1], S. FUJISAWA[2];
[1]Riken Ctr. for Brain Science: RIKEN Noshinkei Kagaku Kenkyu Ctr., Wako/Saitama, Japan;
[2]RIKEN Ctr. For Brain Sci., Wako, Japan

Abstract: Episodic memory is long-term memory that allows us to recall specific events or episodes from our personal experiences. Understanding the neural mechanisms of episodic memory is an essential challenge for neuroscience. However, the neural mechanisms of episodic memory are not well understood. The question is how information about events experienced by individuals is encoded in the neural network. We point out that the brain's information encoding of episodic memory is similar to the holographic principle, which is the most promising candidate for the quantum gravity theory in physics. Quantum gravity theory is a theory that aims to unify Einstein's general relativity and quantum mechanics. The holographic principle originates in black holes, the solution to general relativity. It is theoretically derived that event information sucked into a black hole is encoded not in the three-dimensional space inside the black hole but in the two-dimensional space on the surface of the black hole. This encoding of information in n-dimensional space into n-1-dimensional space, one dimension lower, is called the holography principle. In recent years, many studies in physics have pointed out that the process of emergence of n-dimensional space from n-1-dimensional space in the holography principle is similar to neural networks. We point out that encoding event information in black holes has many similarities to encoding events in the hippocampal network which is essential for episodic memory, and propose a new model of episodic memory based on similarities to the holographic principle. We show that the Einstein equations describing the dynamics of black hole spacetime can be derived from neural networks. Our model predicts that when encoding event information in the hippocampal network, encoding hierarchical structure of events and chaos will appear, as seen in black holes. We also propose some ideas for testing these predictions in animal experiments.

Disclosures: D. Kawahara: None. **S. Fujisawa:** None.

Poster

PSTR170. Episodic and Episodic-Like Memory-Neurophysiology and Models

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR170.13/PP23

Topic: H.07. Long-Term Memory

Support: University of Saskatchewan College of Medicine
Natural Sciences and Engineering Research Council of Canada

Title: The anterior retrosplenial cortex is required for long-term object-in-place associative memory retrieval: role of ionotropic glutamate receptors in male and female Long Evans rats

Authors: *D. L. MCELROY, Q. GREBA, H. SABIR, A. E. GLASS, J. G. HOWLAND; Anatomy, Physiology, and Pharmacol., Univ. of Saskatchewan, Saskatoon, SK, Canada

Abstract: The anterior retrosplenial cortex (aRSC) integrates multimodal sensory information into cohesive associative memories. Little is known about how information is integrated at different learning phases (i.e., encoding and retrieval). Additionally, sex differences are observed in some visuospatial memory tasks; however, inconsistent findings warrant more research. The following experiments used the object-in-place (OiP) test to assess associative memory. Briefly, rats explore four novel objects in a box for 5 minutes (sample), return to their home cage for 60 minutes (delay), then return to the box for 4 minutes to explore an identical copy of the sample objects, but two objects have swapped locations (test). Given rats reliably exhibit a preference towards novelty, enhanced exploration of the novel object-location pairs is interpreted as intact associative memory. Three experiments were conducted using the OiP test and 1-hour delay: (i) First, we assessed sex differences in OiP using naive male (n=12) and female (n=12) Long Evans rats. Results show females perform equally well in OiP, suggesting carefully controlling stressors and thoroughly habituating rats (7 days) may be sufficient to eliminate reported sex differences. (ii) Next, we determined the role of the aRSC in long-term OiP *retrieval*. To this end, bilateral cannulae were surgically implanted into the aRSC of male (n=11) and female (n=11) Long Evans rats prior to testing. Following recovery, rodents were tested on OiP and a mixture of muscimol/baclofen ($GABA_{A/B}$ receptor agonists) or saline (0.9%) was infused ~15 minutes prior to the test phase, temporarily lesioning the aRSC. Results show that aRSC lesions significantly impaired OiP performance in both sexes ($p<0.05$). (iii) Currently, we are assessing the role of aRSC ionotropic glutamate receptors in OiP retrieval, using cannulated male (n=9) and female (n=8) Long Evans rats. The design is identical to (ii), except that infusions include the competitive NMDA receptor antagonist AP-5 (30 mM), competitive AMPA/Kainate receptor antagonist CNQX (3 mM), or saline (all volumes = 0.50 uL/side). Preliminary data suggest AP-5 impairs long-term OiP retrieval ($p < 0.05$), while CNQX results are not yet interpretable. Taken together, these findings challenge reported sex differences and clearly establish the aRSC in long-term associative memory retrieval. Preliminary findings also suggest that ionotropic glutamate receptor activation in aRSC is involved in long-term retrieval. Thus, modulating neural activity in aRSC may alleviate associative memory impairments in those with brain disorders.

Disclosures: D.L. McElroy: None. **Q. Greba:** None. **H. Sabir:** None. **A.E. Glass:** None. **J.G. Howland:** None.

Poster

PSTR170. Episodic and Episodic-Like Memory-Neurophysiology and Models

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR170.14/PP24

Topic: H.07. Long-Term Memory

Support: NIH GRANT R01 MH 119099

Title: Quantifying persistent mental content

Authors: *G. KRESSIN PALACIOS[1], B. BELLANA[2], C. J. HONEY[1];
[1]Johns Hopkins Univ., Baltimore, MD; [2]York Univ., Baltimore, ON, Canada

Abstract: In the study of human memory, we often focus on how memories enable us to achieve specific goals, such as locating our phone or wallet. However, another form of memory is the persistence of experiences in our minds over minutes and hours without overt volition. This persistence can be beneficial, enabling us to find creative solutions to problems, but it can also be detrimental, leading to repetitive unwanted thoughts that are associated with depression and anxiety. How can we quantify the persistence of recent experiences in ongoing thought? Bellana et al. (2022) introduced a paradigm to assay spontaneous thought using word chains generated by individuals before and after reading an immersive story. Participants' word-chains contained reliable semantic biases related to the story. Moreover, they reported lingering thoughts about the story after it ended. We hypothesized that we could use each participant's (objective) word-chain biases to predict their (subjective) report of lingering thoughts. To this end, we scored over 7,000 words on their story-relatedness. These word norms significantly improved our capability to detect semantic biases in word chains, relative to previous methods using word embeddings. Using our word norms, we found that participants with stronger story-related semantic biases also reported stronger lingering of the story in mind. Furthermore, the semantic biases persisted for at least 5 minutes beyond the end of the story. Finally, these biases were robust even in participants whose lingering thoughts were non-volitional. Altogether, we enhanced our tools for objectively quantifying content that persists in human thought without overt volition.

Disclosures: G. Kressin Palacios: None. **B. Bellana:** None. **C.J. Honey:** None.

Poster

PSTR170. Episodic and Episodic-Like Memory-Neurophysiology and Models

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR170.15/PP25

Topic: H.07. Long-Term Memory

Support: Swebilius Foundation

Title: Modulation of pattern separation in episodic memory by direct electrical stimulation of the human hippocampus

Authors: *I. ZHOU[1], K. N. GRAVES[1], E. A. MCDEVITT[4], G. L. STEMERMAN[1], K. L. GEISEL[1], K. A. NORMAN[4,5], N. B. TURK-BROWNE[1,2], I. H. QURAISHI[3];
[1]Psychology, [2]Wu Tsai Inst., [3]Neurol., Yale Univ., New Haven, CT; [4]Princeton Neurosci. Inst., [5]Psychology, Princeton Univ., Princeton, NJ

Abstract: The hippocampus is critically involved in episodic memory. Much of our understanding of this involvement is based on animal models, human lesion studies, and human

brain imaging. Invasive neurostimulation offers another causal test of the necessity of the human hippocampus for episodic memory, with key advantages of being direct, highly localized, and reversible. Although hippocampal stimulation has been explored in patients undergoing acute intracranial evaluations of intractable epilepsy, its effects on memory remain unclear, with conflicting evidence supporting enhancement, impairment, and null effects. Here, we perform direct hippocampal electrical stimulation in adults with epilepsy previously implanted with chronic responsive neurostimulation (RNS) devices. We aimed to disentangle the divergent effects reported in prior studies and to unpack the role of the hippocampus in specific episodic memory computations. In particular, we focus on pattern separation, a hippocampal process that differentiates representations of similar experiences to prevent interference. We hypothesized that 1000-ms bursts of high-frequency hippocampal stimulation would elicit reliable impairment of pattern separation. The task consisted of three phases. In the first phase, participants engaged in incidental encoding of object images. This was immediately followed by two blocks of a test phase — one with stimulation and one without, order counterbalanced — in which participants saw target, lure, and foil images and indicated whether they were "old", "similar", or "new". The ability to identify lure images as "similar" rather than "old" is a behavioral marker of pattern separation. Participants then engaged in a post-test phase in which they viewed lure items from the test phases and their original counterparts from the encoding phase to examine neural differentiation of their representations. Local field potentials were recorded from RNS contacts in all non-stimulation phases. Patients with unilateral or bilateral hippocampal contacts received bipolar stimulation exclusively to the hippocampal contacts, which were validated via localization of electrodes in pre- and post-implant anatomical images. Additional control patients received stimulation outside of the hippocampus. Preliminary results suggest that pattern separation behavior may be selectively impaired by hippocampal stimulation, although testing is ongoing. These findings suggest a promising direction for direct manipulation of precise circuit functions using implanted neurostimulators, allowing for future causal investigations of hippocampal function.

Disclosures: **I. Zhou:** None. **K.N. Graves:** None. **E.A. McDevitt:** None. **G.L. Stemerman:** None. **K.L. Geisel:** None. **K.A. Norman:** None. **N.B. Turk-Browne:** None. **I.H. Quraishi:** None.

Poster

PSTR170. Episodic and Episodic-Like Memory-Neurophysiology and Models

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR170.16/PP26

Topic: H.07. Long-Term Memory

Support: NIA Grant 1R01AG053961

Title: Higher number of biological children is associated with increased risk of cognitive dysfunction in older African American women

Authors: S. M. ABEDULLAH, Z. OSIECKA, B. A. FAUSTO, *M. A. GLUCK;
Ctr. for Mol. and Behavioral Neurosci., Rutgers University–Newark, Newark, NJ

Abstract: Higher number of biological children is associated with increased risk of cognitive dysfunction in older African American women
Authors Salma M. Abedullah, Zuzanna Osiecka, Bernadette A. Fausto, Ph.D., & Mark A. Gluck, Ph.D.
African Americans are two to three times more likely to develop Alzheimer's disease (AD). Studies show that women have a higher prevalence of AD than men--putting older African American women at a disproportionate risk. Previous research demonstrates that the number of children a woman has had is associated with AD neuropathology. This disparity may be attributed to differences in hormonal patterns following childbirth. The objective of this project is to compare hippocampal-dependent cognitive performance between older African American women with and without children and to explore the interactive effects of socioeconomic status (SES) and the number of biological children on overall cognitive performance. The study sample consisted of 100 cognitively healthy African American women aged 60 and older recruited from the greater Newark area. Participants reported the number of biological children, education level, age, depressive symptomology as measured by the Geriatric Depression Scale, and median household income. Participants' cognitive performance was assessed using the Rey Auditory Verbal Learning Test (RAVLT) and a Rutgers Generalization Task (Concurrent Discrimination and Transfer Task). Results show that the number of biological children a participant has had is significantly associated with performance on the generalization task but not on RAVLT. As the number of children increases, the number of errors made on the generalization task also increases. Generalization tasks may better assess early changes in cognitive performance in older African American women who have had children than standard episodic memory assessments. The relationship between estrogen depletion and the number of biological children may shed light on the mechanism by which having children is associated with poorer cognitive function. Future studies should explore how these findings can be applied to protecting cognitive function in older African American women who have had biological children.

Disclosures: S.M. Abedullah: None. **Z. Osiecka:** None. **B.A. Fausto:** None. **M.A. Gluck:** None.

Poster

PSTR170. Episodic and Episodic-Like Memory-Neurophysiology and Models

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR170.17/PP27

Topic: H.07. Long-Term Memory

Title: Differential engagement of dorsal and ventral medial prefrontal cortex in retrieval of episodic and semantic memory

Authors: *S. D. ALLEN[1], C. G. CONNOLLY[2], C. B. MARTIN[1];
[1]Psychology, [2]Col. of Med., Florida State Univ., Tallahassee, FL

Abstract: The distinction between semantic and episodic memory as two separate long-term memory systems was first proposed in 1972 by Endel Tulving. These systems are primarily characterized by differences at the level of content; semantic memory reflects knowledge abstracted from multiple experiences whereas episodic memory reflects memory for spatiotemporally unique moments. In most cognitive neuroscience research, semantic and episodic memory are probed in independent experiments. As such, questions remain regarding the extent to which these fundamentally different types of retrieval engage similar vs. different cortical areas. Relevant neuroimaging studies and research in patients has revealed perirhinal cortex as an area that makes critical contributions to semantic and episodic retrieval when using objects or object concepts as stimuli. It is unknown, however, whether retrieval of task relevant information from this structure engages different frontally-mediated control networks. We addressed this question using functional magnetic resonance imaging (fMRI) in thirty cognitively healthy human participants. During scanning, participants processed each object concept semantically (i.e., natural/manmade) in one task context and episodically (i.e., old/new) in a separate task context. We used a group-level analysis to reveal a functional distinction between the dorsal and ventral extents of medial prefrontal cortex (MPFC). Specifically, ventral MPFC was differentially active during retrieval of semantic memory, whereas dorsal MPFC was differentially active during retrieval of episodic memory. This pattern of results reveals a role for MPFC in both types of retrieval with a functional distinction along its dorsal and ventral extents. More broadly, this finding is consistent with evidence from neuroimaging research that has revealed a dorsal-to-ventral hierarchy of cognitive control in lateral prefrontal cortex in other task contexts. This functional gradient is thought to be captured by differences in degrees of abstraction, with more ventral/anterior regions supporting more abstract forms of control.

Disclosures: S.D. Allen: None. **C.G. Connolly:** None. **C.B. Martin:** None.

Poster

PSTR170. Episodic and Episodic-Like Memory-Neurophysiology and Models

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR170.18/Web Only

Topic: H.07. Long-Term Memory

Support: UNAM DGAPA PAPIIT IG300121
CONAHCYT 997227

Title: Electrophysiological activity distinctions between spatial and temporal context in episodic memory

Authors: *C. TORRES-MORALES[1], S. CANSINO[2];
[1]Lab. of NeuroCognition, Univ. Nacional Autónoma de México, Mexico City, Mexico, Mexico

City, Mexico; [2]Lab. NeuroCognition, Nat Autonomous Univ. of Mexico, Univ. Nac Autónoma de México, Mexico City, Mexico

Abstract: Episodic memory refers to our ability to mentally retain personal experiences that occurred in a particular spatial and temporal context. Brain activity distinctions for spatial and temporal context have been observed in nonhuman and humans through several techniques, such as unicellular electrical recording and functional magnetic resonance imaging. However, these distinctions have not been examined through electrophysiological recordings of neural populations such as event-related potentials (ERP) because memory for spatial and temporal context has not been assessed together in a same ERP study. The study aimed to examine electrophysiological activity distinctions between spatial and temporal context during encoding and retrieval. ERPs were recorded in 30 participants while performing an associative task. During encoding and retrieval, pairs of images were presented sequentially. In the retrieval task, participants identified four possible trials: the order and location of the images were identical to those presented during encoding; the images were presented in a different order; the images were presented in different locations; or the images were new. Participants' performance was equivalent for spatial and temporal context retrieval. Subsequent memory for correct temporal context, compared to that for correct spatial context, elicited during encoding a larger positive component between 830 and 1060 ms in frontal derivations after the onset of the second image. Correct temporal context retrieval elicited greater amplitude waveforms than correct spatial context retrieval between 300 and 450 ms and 500-850 ms after the onset of the second image. The early component was observed in the right and middle frontal sites and the middle central sites. The differences in the latter component were observed across frontal, central, and parietal sites. The findings revealed that the encoding and retrieval of temporal context in episodic memory is associated with greater amplitude waveforms than spatial context, even when the difficulty in processing each context was equivalent.

Disclosures: C. Torres-Morales: None. **S. Cansino:** None.

Poster

PSTR170. Episodic and Episodic-Like Memory-Neurophysiology and Models

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR170.19/PP28

Topic: H.07. Long-Term Memory

Support: NRF of Korea, Basic Science Research Program (2020R1A2C2007770)
NRF of Korea, Neurological Disorder Research Program (2020M3E5D9079913)
SNU, New Faculty Startup Fund

Title: A common process of self-distancing during memory retrieval across diverse memory contents

Authors: *I. SHIN[1,2], S.-H. LEE[2];
[1]Korea Advanced Inst. of Sci. and Technol., Daejeon, Korea, Republic of; [2]Dept. of Psychology, Seoul Natl. Univ., Seoul, Korea, Republic of

Abstract: Autobiographical memories are typically retrieved from a first-person perspective, but individuals can also retrieve memories from the viewpoint of a distant observer by taking psychological distance. This cognitive process, known as self-distancing, stands in contrast to self-immersion. Previous research has highlighted the benefits of self-distancing in regulating negative emotions during the retrieval of negative memories. However, the neural processing underlying self-distancing during memory retrieval remains poorly understood. We performed a functional magnetic resonance imaging (fMRI) experiment to reveal the neural bases of self-distancing during retrieval. The experiment was composed of a pre-scan interview session and a retrieval session. During the pre-scan interview session, vivid autobiographical memories containing various contents were collected from each participant. During the retrieval session, they retrieved the memories collected during pre-scan interview session inside the MRI scanner. They performed the retrieval task under two conditions: self-distancing, where they retrieved memories from a distant perspective, and self-immersion, where they retrieved memories from their own perspective. The mean activation level in the prefrontal and parietal regions was significantly higher during the self-distancing condition compared to the self-immersion condition. Significant decoding of memory contents was not observed throughout the entire brain during the self-distancing condition, while it was possible during the self-immersion condition. Furthermore, significantly stronger shared neural representations were found across the trials using the self-distancing perspective in the visual cortex, dorsolateral prefrontal cortex, and angular gyrus, compared to the trials of the self-immersion condition. These results suggest that memory retrieval with self-distancing involves common neural processes across various memory contents in the visual cortex, dorsolateral prefrontal cortex, and angular gyrus.

Disclosures: I. Shin: None. **S. Lee:** None.

Poster

PSTR170. Episodic and Episodic-Like Memory-Neurophysiology and Models

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR170.20/QQ1

Topic: H.07. Long-Term Memory

Title: Beneficial effects of 5-HT$_4$Rs activation in mice: a transversal approach, from memory to its hippocampal correlates

Authors: *C. ROUX[1], ***C. ROUX**[2], E. ESNEAULT[3], M. LÉGER[2], T. FRERET[2];
[1]COMETE INSERM/UNICAEN U1075, Caen, France; [2]UNIVERSITE DE CAEN / INSERM U1075, Caen, France; [3]Porsolt, Porsolt, Le Genest St Isle, France

Abstract: Beneficial effects of 5-HT$_4$Rs activation in mice: a transversal approach, from memory to its hippocampal correlates

Candice M Roux [1,2], Elise Esneault [2],, Marianne Leger [1], and Thomas Freret [1][1]UNICAEN, INSERM, COMETE, CYCERON, Normandie University, Caen, 14000 Caen, France; [2]PORSOLT, 53940 Le Genest Saint-Isle, France; eesneault@porsolt.com

Abstract

Type 4 serotonin receptors (5-HT$_4$Rs) have earned a place in the sun as a promising therapeutic target for the treatment of memory disorders. Indeed, both pro-mnesiant and anti-amnesiant effects of 5-HT$_4$Rs activation have been repeatedly described in rodents and more recently pro-cognitive effects were identified in healthy volunteers. Despite some hypotheses have been raised, mechanisms at works still remain to be elucidated. A better understanding of the underpinning mechanisms would help to extend the beneficial effects of pharmacological 5-HT$_4$Rs stimulation - *so far limited to the fields of Alzheimer's and Major depressive disorders as central nervous system diseases* – to additional brain pathologies such as Parkinson's and Schizophrenia. These disorders are characterized by early decline in episodic memory that are associated with alterations of hippocampal functioning. Hence, we herein addressed such mechanistic issue through a transversal approach. We investigated the effects of systemic administration of the 5-HT$_4$Rs agonist RS67333 on different functions of hippocampal-dependent episodic-like memory and its neurobiological correlates such as hippocampal synaptic plasticity as well as plasticity-related brain oscillations and neurotransmitters. We identified location and novelty discrimination as two domains of episodic memory that could benefit from 5-HT$_4$Rs activation. Besides, while hippocampal theta power was increased, the magnitude of long-term potentiation was reduced in a frequency-dependent manner. These changes were accompanied by reduced levels of excitatory neurotransmitter glutamate in the hippocampus. Overall, our results support that the beneficial effects of 5-HT$_4$Rs activation on memory are intimately linked to changes in hippocampal synaptic plasticity. The latter are likely due to the observed variations in neurotransmitter levels and dependent oscillatory rhythms that are relevant for plasticity processes.

Disclosures: C. Roux: None. **C. Roux:** None. **E. Esneault:** None. **M. Léger:** None. **T. Freret:** None.

Poster

PSTR170. Episodic and Episodic-Like Memory-Neurophysiology and Models

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR170.21/QQ2

Topic: H.07. Long-Term Memory

Support: European Molecular Biology Organization nonstipendiary Long-Term Fellowship (848–2017)
Human Frontier Science Program (LT000444/2018)
the European Union's Horizon 2020 research and innovation programme under the Marie Skłodowska-Curie Grant Agreement No. 789040

Change is Key! of the Riksbankens Jubileumsfond (M21-0021)
Wellcome collaborator award (214314/Z/18/Z)
Wellcome Trust Senior Research Fellowship (104765/Z/14/Z)
Principal Research Fellowship (219525/Z/19/Z)
James S. McDonnell Foundation Award (JSMF220020372)
Israeli National Postdoctoral Award Program for Advancing Women in Science
Blavatnik Postdoctoral Fellowship

Title: The human brain reactivates neural representations of remote past events to comprehend unfolding experiences

Authors: *A. HAHAMY-DUBOSSARSKY[1], H. DUBOSSARSKY[2], T. BEHRENS[3];
[1]Wellcome Trust Ctr. For Neuroimaging, London, United Kingdom; [2]Queen Mary Univ., London, United Kingdom; [3]Wellcome Ctr. for Integrative Neuroimaging, Univ. of Oxford, Oxford, United Kingdom

Abstract: How will your brain make sense of this abstract? As in other real-life experiences, your brain now encounters a continuous flow of information (e.g. words), but it chunks this flow into discrete "events" (e.g. sentences, Zacks et al., 2007). But to understand an unfolding experience, links must be drawn between the current event and relevant past events. For instance, to understand the conclusions of this abstract (which will be provided in the final sentence), these conclusions must be linked to the research question (which will appear now): How are these links between relevant events formed? Spatial navigation studies conducted on rodents propose that offline neural reactivations ("replay") can tie together several past events, even remote ones, as if building a model of the environment. Here we hypothesized that a similar mechanism could underlie the human ability to bind naturalistic information across time, forming an internal representation of unfolding experience. To test this hypothesis, we developed a new method for detecting reactivation of temporally-remote naturalistic events in human fMRI data. We applied this method to analyse two independent datasets of participants who were watching a movie (n=17, 10 female, ages 18-26), or listening to an audio story (n=25, 14 female, ages 18-40) while beings scanned. We discovered that humans indeed reactivate neural representations of remote past events. Similar to offline replay in rodents, these reactivations occurred in hippocampus, Angular gyrus and precuneus, where only events that are specifically relevant for understanding the current narrative stage were reactivated. However, unlike in rodents, these reactivations occurred at the transitions between ongoing narrative events, rather than during prolonged offline periods. Of importance, these findings were replicated across the two independent datasets, which not only highlights their robustness, but also indicates their generalizability across different types of naturalistic experiences (movie-watching, story-listening). We therefore propose reactivations as a candidate mechanism for binding temporally distant information into a coherent understanding of ongoing experience (such as this abstract!).

Disclosures: A. Hahamy-Dubossarsky: None. **H. Dubossarsky:** None. **T. Behrens:** None.

Poster

PSTR170. Episodic and Episodic-Like Memory-Neurophysiology and Models

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR170.22/QQ3

Topic: H.07. Long-Term Memory

Support: VR #201703135
Advanced ERC Grant SELF-UNITY #787386

Title: Own Body Perception in the Encoding and Retrieval of Naturalistic Events

Authors: *H. IRIYE, H. H. EHRSSON;
Karolinska Inst., Solna, Sweden

Abstract: The ability to perceive one's body as one's own (i.e., body ownership) is crucial to memory. Disrupting own-body perception during encoding reduces memory accuracy and re-experiencing during retrieval. Yet, the underlying mechanism explaining the impact of own-body perception on memory is unclear. Own-body perception, typically a constant feature of encoding and retrieval, may act as a fundamental contextual memory cue. Memory is context dependent, such that retrieval benefits when a memory is recalled in the same context as encoding. Own-body perception may be an especially strong cue given its high self-relevance. We immersed healthy participants (N = 27) within pre-recorded videos seen through virtual reality glasses, which depicted naturalistic events that included a first-person view of a mannequin's reclining body aligned with participants' real bodies during fMRI scanning. Participants saw an object touch the mannequin and simultaneously felt touches on the corresponding location of their real body, which created an illusory sense of ownership over the mannequin. As a control condition, we disrupted the illusion by delivering seen and felt touches asynchronously in half of the videos. 1 week later, participants were re-immersed within videos depicting the mannequin receiving dynamic visuotactile stimulation overlaid on still-frames of the previously seen videos during fMRI scanning. We again manipulated feelings of ownership over the mannequin, which was either congruent or incongruent with how a video was encoded, while participants retrieved memories for the videos. Participants answered cued recall questions concerning specific event details at the end of each session to assess memory accuracy. A linear mixed model showed that a strong sense of body ownership during encoding, but not at retrieval, decreased memory accuracy over the week delay (p = .029), which was not present when encoding and retrieval contexts matched or when body ownership was weak during encoding. Preliminary fMRI results suggest that patterns of activity in medial prefrontal cortex at retrieval discriminate between memories according to their encoding and retrieval context (p = .026, $M_{\text{Decoding Accuracy}}$ = 27.16%, SD = 4.75%). A t-test revealed higher decoding accuracy when memories were both encoded and retrieved with a strong sense of body ownership, compared to when it was strong during encoding but weak during retrieval (p = .005). Collectively, these initial findings imply that a stable sense of body ownership during encoding acts as a contextual memory cue during retrieval, which calibrates patterns of activity in medial prefrontal regions to support remembering.

Disclosures: H. Iriye: None. **H.H. Ehrsson:** None.

Poster

PSTR171. Hippocampal-Cortical Interactions I

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR171.01/QQ4

Topic: H.08. Learning and Memory

Support: CIHR Project Grant #367017
CIHR Project Grant #377074
CIHR Project Grant #463403
NSERC Discovery Grant # 74105
Canada Research Chairs Program

Title: Temporal difference learning theory predicts the evolution of hippocampal neuronal dynamics during learning a reward-based navigation task

Authors: *M. YAGHOUBI[1,2], A. NIETO-POSADAS[1], C.-A. MOSSER[1], É. WILSON[1], T. GISIGER[1], S. WILLIAMS[1,2], M. P. BRANDON[1,2];
[1]Douglas Mental Hlth. University, McGill Univ., Montreal, QC, Canada; [2]Integrated Program in Neuroscience, McGill University,, Montrea, QC, Canada

Abstract: The hippocampus plays a crucial role in constructing a cognitive map of the environment, which aids in navigation and memory-dependent behaviors. Recent work has revealed that this spatial representation undergoes important changes when reward locations are learned. For instance, place fields tend to concentrate near reward locations, and remap when reward contingencies are changed. Here, we aim to understand the emergence and evolution of this reward-related representation across weeks as mice learn a hippocampal-dependent memory task. We employed one-photon miniscopes to image the activity of large ensembles of CA1 neurons during a Delayed Nonmatching-to-Sample touchscreen task. In the sample phase of this task, a white square is presented in one of five positions on the touchscreen. A nose poke to this square starts a delay period (increasing from 2 to 8 seconds as the mice learn the task). Following this delay, two white squares are displayed, and the mouse must choose the nonmatching square to receive the reward, located on the back wall of the touchscreen chamber. Analysis of these data reveals the following: 1) A remarkably precise representation of location (mean decoding error ~4cm). 2) As animals learn the task, distinct neuronal responses emerge. First, place cells become directionally tuned. Second, a subset of the population shifts from encoding location to encoding either the presence of a touchscreen cue, reward, or reward prediction. ‘Reward prediction cells’ encode the value of the reward through the amplitude of their neural responses. 3) Across weeks, as mice master the task, the proportions of these specialized cell types change in accordance with that expected by the Temporal Difference Learning theory. The proportion of reward cells decreases while the proportion of cue-selective and reward-prediction cells both increase. Together, these results describe how CA1 populations evolve over time as animals master a hippocampal-dependent memory task and offer support to the Temporal Difference Learning theory.

Disclosures: **M. Yaghoubi:** None. **A. Nieto-Posadas:** None. **C. Mosser:** None. **É. Wilson:** None. **T. Gisiger:** None. **S. Williams:** None. **M.P. Brandon:** None.

Poster

PSTR171. Hippocampal-Cortical Interactions I

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR171.02/QQ5

Topic: H.08. Learning and Memory

Support: CIHR Project Grant #367017
CIHR Project Grant #377074
CIHR Project Grant #463403
NSERC Discovery Grant #74105
Canada Research Chairs Program
CIHR Postdoctoral Fellowship #472750

Title: Population dynamics of CA1 spatial coding during drift and reorientation.

Authors: *J. LEE[1], T. XU[2], M. P. BRANDON[3];
[1]Psychiatry, McGill Univ., Montreal, QC, Canada; [3]McGill Univ., [2]McGill Univ., Montréal, QC, Canada

Abstract: To flexibly navigate environments the brain must continuously update its sense of direction and position with visual landmarks and self-motion cues. Prior work has shown that movement of visual landmarks is sufficient to drive reorientation in the brain's mapping of direction, position, and even self-motion. Recent work on population coding of head direction in the anterior-dorsal thalamic nucleus (ADN) has further shown that average network activity, termed network gain, constrains the dynamics of reorientation during movement of a visual landmark, and short-term drift in its absence. Indeed, modelling of simple attractor networks predicts that network gain is a general constraint for representational stability and updating of arbitrary neural codes. Given the pervasiveness of attractor-like neural architectures throughout the brain, network gain could determine the dynamics of representational stability and updating in neural systems beyond the ADN. Here we record large neural populations in CA1 of freely-behaving mice in augmented reality while a visual landmark is manipulated instantaneously to induce reorientation and drift of self-position coding. We compare these results to simple attractor network models, and the observed relationship between network gain and reorientation in CA1 against prior observation in ADN. The similarities and differences across these regions and models, which encode distinct behavioural latents at the population-level (direction vs. position), will help uncover how population-level dynamics determine representational stability and updating across diverse neural systems.

Disclosures: **J. Lee:** None. **T. Xu:** None. **M.P. Brandon:** None.

Poster

PSTR171. Hippocampal-Cortical Interactions I

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR171.03/QQ6

Topic: H.08. Learning and Memory

Support: R34NS123819 (NIH/NINDS)

Title: A Bayesian inference model predicts the head direction system's response to changing visual information

Authors: *Z. AJABI[1], J. DRUGOWITSCH[2];
[1]Harvard Univ., Boston, MA; [2]Dept. of Neurobio., Harvard Med. Sch., Boston, MA

Abstract: Animals use a variety of sensory modalities (e.g. visual, olfactory, auditory, proprioceptive, etc⋯) to navigate space. The reliability of sensory information may vary across modalities, such that the brain ought to prioritize certain inputs over others or combine them in some principled way. Bayesian inference provides such a principled approach, but whether the nervous system uses Bayesian inference for navigation remains an open question. The head direction system provides an ideal basis to address this question. Specifically, we asked how the HD system ought to learn and re-learn the associations between HD and visual scenes. The HD system is known to be highly reliant on visual information. Indeed, studies in both vertebrates and invertebrates showed that the internal HD representation appear to frequently shift and realign with changing visual reference frames irrespective of self-motion or other non-visual external sensory information - a phenomenon known as 'reset'. These resets do not only vary in speed, but are also sometimes absent, leading to an unchanged internal HD representation despite a changed visual reference frame - a phenomenon known as 'remap'. We asked if the brain arbitration between remaps and resets, and the speed of these resets, could be explained by the brain continuously solving a causal inference problem to determine whether a visual context change has occurred or not. To do so, we designed a Bayesian model that performs evidence accumulation to infer context changes which, in turn, determines the reset speed. We found that the model's response to changes in the visual input can be fully predicted from a learning rate that reflects the animal's expected context switch rate, as well as the reliability of new visual observations. Highly reliable visual observations favor learning new HD-visual associations (i.e. remap) while low learning rates promote maintaining old associations (i.e. reset). Interestingly, our model can maintain a memory trace of the orientation of previously displayed visual landmarks for a duration that depends on the aforementioned parameters, in agreement with recent experimental studies. Finally, we explore the behavioral and experiential factors that ought to impact the learning rate, based upon which we derive the learning rule for optimal navigation strategies promoting stable internal HD representation.

Disclosures: Z. Ajabi: None. **J. Drugowitsch:** None.

Poster

PSTR171. Hippocampal-Cortical Interactions I

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR171.04/QQ7

Topic: H.08. Learning and Memory

Support: NIH R01MH132171

Title: The time of future action is represented with continuous time constants in mPFC

Authors: *R. CAO, I. M. BRIGHT, M. W. HOWARD;
Psychological and Brain Sci., Boston Univ., Boston, MA

Abstract: It is believed that we remember the past in order to predict the future. Findings from entorhinal cortex demonstrate that the timing of a past event can be decoded from "temporal context cells". Immediately after the event, the firing of a population of temporal context cells is simultaneously perturbed and then relaxes back to baseline exponentially at a spectrum of rates. Because the rate constants are distributed smoothly, the firing of this population approximates the Laplace transform of the time of the remembered event in the past. In the interval reproduction task, the animal must remember the event starting the interval and anticipate the time of the planned response to terminate the interval, allowing us to study both memory for the past and anticipation of the future. We analyzed previously published recordings from mPFC during the interval production period using hierarchical Bayesian modeling of temporal receptive fields. We identified two groups of cells that represent the time of events. One group of cells represents the time since the event that began the interval; this population resembled temporal context cells. Another group ramped in anticipation of the action that terminates the delay, peaking at the end of the interval. Critically, the firing of these ramping neurons also changed at a variety of rates, mirroring the pattern of the "temporal context cells". The hierarchical Bayesian analysis provided strong evidence for a power law distribution of rate constants, falsifying the hypothesis that there is a characteristic scale to the rate of anticipatory ramps. The firing of the population of ramp cells thus approximates the Laplace transform of the time of anticipated future events.

Disclosures: R. Cao: None. **I.M. Bright:** None. **M.W. Howard:** None.

Poster

PSTR171. Hippocampal-Cortical Interactions I

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR171.05/QQ8

Topic: H.08. Learning and Memory

Support: CIHR Project Grant #367017
CIHR Project Grant #377074
CIHR Project Grant #463403
NSERC Discovery Grant #74105
Canada Research Chairs Program
Vanier Canada Graduate Scholarship

Title: Hippocampal modulation of dopamine prediction error signalling

Authors: *E. J. P. MAES[1], M. P. BRANDON[2];
[2]Psychiatry, [1]McGill Univ., Montreal, QC, Canada

Abstract: The phasic release of dopamine (DA) from midbrain neurons has been implicated in the encoding of prediction error, as well as salience, to drive learning. Similarly, hippocampus is a crucial region in the encoding of memory. Moreover, stimulation of CA1/Subiculum has been shown to elicit strong responses in DA neurons. However, there is a lack of evidence for the exact role of hippocampus in regulating DA release, and its impact on memory encoding. We addressed this question using chemogenetics and fiber photometry in freely-moving mice on a linear track. We recorded dopamine release in the NAc using GRAB-DA2m fiber photometry, coupled with ventral hippocampal disinhibition using DREADDs. We found that mice adapted shuttling behavior according to the probability of upcoming reward, and that DA release conformed to reward prediction error based on anticipated reward probability. On test day, vCA1/vSub was disinhibited via DREADD inhibition of PV+ GABAergic interneurons to determine the role of hippocampus in regulating DA prediction error computation. Revealing how the hippocampus and DA orchestrate prediction and learning is fundamental to understanding the basis of behavior, as well as disorders such as schizophrenia, where patients show a combination of dysregulation of hippocampus, hyperactive DA release, and cognitive impairments.

Disclosures: E.J.P. Maes: None. **M.P. Brandon:** None.

Poster

PSTR171. Hippocampal-Cortical Interactions I

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR171.06/QQ9

Topic: H.08. Learning and Memory

Support: NIH Grant MH120073
NIH Grant MH060013
ONR MURI N00014-19-1-2571

Title: Distinct codes for environmental structure and symmetry in postrhinal and retrosplenial cortices

Authors: *P. A. LACHANCE, M. E. HASSELMO;
Psychological and Brain Sci., Boston Univ., Boston, MA

Abstract: The physical boundaries of an enclosed space provide cues for establishing one's spatial orientation. For example, an animal's heading can be measured relative to the orientations of nearby flat walls or corners. In a square environment, this signal would be expected to show four-fold radial symmetry, as the environment's boundary structure is identical across 90° rotations. Alternatively, heading can be measured relative to the centroid of the environment (i.e., a simultaneous representation of all boundaries), which should instead result in a circularly symmetric signal. The measurement of heading with respect to specific reference points is called 'egocentric bearing.' To probe which spatial coding scheme is employed by cortical regions implicated in egocentric spatial processing, we simultaneously recorded single neurons from the postrhinal (POR) and retrosplenial (RSC) cortices as rats foraged in a square enclosure. Both POR and RSC have been previously shown to contain neurons sensitive to the egocentric bearings of structural elements of the environment (center-bearing cells or egocentric boundary cells (EBCs), respectively). We found that RSC EBCs tended to show strong four-fold radial symmetry in their spatial firing preferences, suggesting egocentric tuning to nearby boundary orientations. POR cells almost never showed this four-fold symmetry, implying overall egocentric tuning to the environment center. Changing the square environment into an 'L-shaped' environment broke the four-fold environmental symmetry and accordingly disrupted four-fold symmetry in the RSC egocentric representation, implying that RSC EBCs are sensitive to boundary placement and not environmental symmetry itself. In a related experiment, allocentric HD cells were simultaneously recorded from POR and RSC as a prominent environmental feature (a large white cue card) was duplicated on opposite walls of the enclosure, making it bidirectionally symmetrical. As reported previously, POR HD cells became bidirectionally tuned under this condition. On the other hand, while some RSC HD cells became bidirectional, many did not. Overall, the results of this experiment indicate that, while POR and RSC both contain 'egocentric cells' and 'HD cells,' the specific environmental features represented by these neurons in each brain region are highly distinct.

Disclosures: P.A. LaChance: None. **M.E. Hasselmo:** None.

Poster

PSTR171. Hippocampal-Cortical Interactions I

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR171.07/QQ10

Topic: H.08. Learning and Memory

Support: NIH Grant MH120073
NIH Grant MH060013
ONR MURI N00014-19-1-2571

Title: Longitudinal tracking of spatial responses in the retrosplenial cortex to environmental manipulations

Authors: *S. L. MALMBERG[1], L. C. CARSTENSEN[2], A. ALEXANDER[3], G. MATTESSICH[5], A. P. GODDARD[6], W. G. CHAPMAN[3], M. PATEL[3], K. D. PATEL[3], M. E. HASSELMO[4];
[1]Psychological and Brain Sci., [2]Grad. Program for Neurosci., [4]Psychological & Brain Sci., [3]Boston Univ., Boston, MA; [5]Northeastern Univ., Boston, MA; [6]Rochester Inst. of Technol., Henrietta, NY

Abstract: The retrosplenial cortex (RSC) is involved in cognitive processes including spatial navigation and episodic memory. Understanding the arrangement of familiar and novel barriers and other behavioral obstructions in an environment is fundamental for guiding spatial navigation. Because the RSC receives inputs from motor, memory, and sensory regions, the RSC is positioned to integrate this information to generate appropriate behavioral responses to environmental changes and obstructions. Examining how the RSC responds to current and past barrier positions enhances understanding of neural mechanisms underlying adaptive behavior. The capacity of calcium imaging to track populations of hundreds of neurons over hours and days allows this technique to address questions regarding how neurons respond to inserted barriers based on physical obstruction or as a result of novelty. In this current work, we use 1-photon calcium imaging to record large populations of neurons in freely-moving animals over the course of days in a variety of spatial foraging tasks involving the insertion of different objects and barriers. We report that there is evidence of spatial responses in the RSC specific to inserted barriers in a specific location in the environment, and that these responses can be tracked over time to detect changes in neuronal response due to changes in novelty.

Disclosures: S.L. Malmberg: None. **L.C. Carstensen:** None. **A. Alexander:** None. **G. Mattessich:** None. **A.P. Goddard:** None. **W.G. Chapman:** None. **M. Patel:** None. **K.D. Patel:** None. **M.E. Hasselmo:** None.

Poster

PSTR171. Hippocampal-Cortical Interactions I

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR171.08/QQ11

Topic: H.08. Learning and Memory

Support: CIHR Project Grant #367017
CIHR Project Grant #377074
CIHR Project Grant #463403
NSERC Discovery Grant # 74105
Canada Research Chairs Program
NSERC PGS D

Title: Context-dependent representations in the hippocampus coincide with task knowledge duiring paired-associate learning in mice

Authors: *Z. HAQQEE[1], S. LA ROSA[1], S. WILLIAMS[2], M. P. BRANDON[2];
[1]Integrated Program in Neurosci., [2]Psychiatry, McGill Univ., Montreal, QC, Canada

Abstract: Hippocampal place cells demonstrate context-dependent spatial representations of their environment in subjects that have been well-trained on a navigation task. How these representations evolve with learning is seldom studied, particularly in more complex tasks that are gradually learned over many days or weeks. Using one-photon calcium imaging with microendoscopes (miniscopes), we tracked hundreds of cells in dorsal CA1 of the hippocampus of mice gradually learning a paired-associate learning (PAL) touchscreen task. Using a combination of dimensionality reduction techniques and generalized linear models, we identify specific ensembles of cells that dynamically evolve their tuning to spatial and contextual variables over more than a month of daily training on the touchscreen task, from habituation to overtraining phases of learning. Individual cells rapidly transition between hidden states as the animal learns the task, gradually stabilizing their transition rate over time. Cells became transiently tuned to task context during specific days of learning, developing context-specific place fields that then generalized across contexts gradually as the animal became over-trained on the task. This phenomena correlated negatively with a cell's spatial information score, suggesting that non-place-cells were more likely to be context specific than place cells. The data suggest that learning might transiently promote the formation of context-dependent maps in the hippocampus, which are then consolidated as the subject is well-trained and becomes familiar with the task design.

Disclosures: Z. Haqqee: None. **S. La Rosa:** None. **S. Williams:** None. **M.P. Brandon:** None.

Poster

PSTR171. Hippocampal-Cortical Interactions I

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR171.09/QQ12

Topic: H.08. Learning and Memory

Support: FRQS Postdoctoral fellowship #320805
CIHR Project Grant #367017
CIHR Project Grant #377074
CIHR Project Grant #463403
NSERC Discovery Grant #74105
Canada Research Chairs Program

Title: Emergence of structured hippocampal network activity during learning of a memory-dependent navigation task

Authors: *C.-A. MOSSER[1], M. YAGHOUBI[2], A. NIETO-POSADAS[3], Z. HAQQEE[1], S. WILLIAMS[1], M. P. BRANDON[1];
[1]Psychiatry, McGill Univ., Montréal, QC, Canada; [2]Dept. of Psychiatry, Douglas Mental Hlth. University, McGill Univ., Montreal, QC, Canada; [3]Psychiatry, McGill, Montreal, QC, Canada

Abstract: Recent technical advances enable the recording of large neuronal populations during behavior resulting in increasingly complex datasets. Despite bold and important open-science and data-sharing policies, these datasets across laboratories tend to apply unique data acquisition methods, behavior, and file structures. Discrepancies between protocols present key challenges including the comparison of results between brain regions, laboratories, and species. We have established the McGill-Mouse-Miniscope platform (M3, www.m3platform.org) to combine miniscope calcium imaging with standardized touchscreen-based behavioral testing. Our mission is to curate an open-source and standardized framework for acquiring, analyzing, and accessing high-quality data of the neuronal dynamics that underlie behavior and cognition throughout the brain in mice. Each experiment provides up to 1000 simultaneously recorded neurons from a single brain region over the course of ~3 months as animals initially learn and master the task. We have collected several datasets to highlight the feasibility of this approach, including hippocampal CA1 and CA3 recordings during the delayed trial-unique nonmatching-to-location (TUNL) task. The TUNL task assesses spatial working memory and consists of an encoding phase, a delay phase and a retrieval phase. In this task, a white square is presented in one of five positions on the touchscreen. A nose poke to this square starts a delay period. Following this delay, two white squares are displayed, and the mouse must choose the nonmatching square to receive reward. As expected, in this task, CA1 and CA3 neurons are spatially modulated; however, they are also sensitive to other behavioral and task-related features. We are now examining how this structure emerges in CA1 and CA3 regions during the initial learning of this task, and how network activity restructures when memory load is changed. These data will help to shed light on how large populations of hippocampal neurons encode a complex memory task and will allow for comparison across laboratories and mouse models.

Disclosures: C. Mosser: None. **M. Yaghoubi:** None. **A. Nieto-Posadas:** None. **Z. Haqqee:** None. **S. Williams:** None. **M.P. Brandon:** None.

Poster

PSTR171. Hippocampal-Cortical Interactions I

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR171.10/QQ13

Topic: H.08. Learning and Memory

Support: CONACYT
Canada Research Chair Tier 1
NSERC RGPIN-2020-06717
CIHR FDN-148478

Title: The role of Oriens-lacunosum moleculare cells in memory dynamics through brain oscillations coupling.

Authors: *B. A. CONTRERAS[1], J. E. CARMICHAEL[2], S. WILLIAMS[2];
[1]McGill Univ. Integrated Program in Neurosci., Lasalle, QC, Canada; [2]McGill Univ. Douglas Res. Ctr., Lasalle, QC, Canada

Abstract: Oriens-lacunosum moleculare (OLM) interneurons, a subgroup of somatostatin-positive interneurons, are pivotal modulators of hippocampal network activity, crucially influencing the flow of information. These OLM cells act as gatekeepers, managing the input from the entorhinal cortex (EC) and the Schaffer collateral CA3 input onto CA1 pyramidal neurons. This gating mechanism may help control current sensory stimuli, embodied by fast gamma oscillations (~60-120 Hz) from the EC, and the retrieval of existing memories, supported by slow gamma oscillations (~30-60 Hz) from CA3. Activation of OLM cells lead to inhibition of the distal dendrites of CA1 pyramidal cells, which weakens EC inputs, hence down-regulating fast gamma activity (Leao et al., 2012). Simultaneously, OLM cells may facilitate the influence of CA3 inputs (slow gamma) by inhibiting interneurons that would otherwise dampen the CA3's contribution. Consequently, the aggregate effect of OLM cell activation would tend to favor memory retrieval (slow gamma, CA3) over the processing of immediate sensory inputs (fast gamma, EC). Furthermore, OLM cells exhibit rhythmic bursting behavior that aligns with theta oscillations (~8 Hz), suggesting they may coordinate the intricate balance between theta and gamma oscillations. This interplay, known as phase-amplitude coupling (PAC), may orchestrate the timing of neural activity to optimize information processing. PAC is believed to be vital for encoding and retrieving memories in the hippocampus, anchoring the temporal dynamics of information flow. In essence, OLM interneurons, through their regulation of theta and gamma oscillations and probable involvement in theta-gamma coupling, could be instrumental in modulating hippocampal information processing and memory dynamics.
Our project aims to rigorously test the hypothesis that CA1 OLM interneurons play key roles in the generation of theta and gamma oscillations, as well as in theta-gamma PAC in mice. We used optogenetic tools to silence OLM interneurons during electrophysiological CA1 recordings of theta and gamma oscillations in running and REM sleep episodes. Additionally, we will assess whether manipulating OLM interneurons ontogenetically influences memory consolidation during REM sleep. The data presented here will provide a new perspective on the role of OLM interneurons in memory dynamics.Leão RN, Mikulovic S, Leão KE, Munguba H, Gezelius H, Enjin A, Patra K, Eriksson A, Loew LM, Tort AB, Kullander K. OLM interneurons differentially modulate CA3 and entorhinal inputs to hippocampal CA1 neurons. Nat Neurosci. 2012 Nov;15(11):1524-30. doi: 10.1038/nn.3235.

Disclosures: B.A. Contreras: None. **J.E. Carmichael:** None. **S. Williams:** None.

Poster

PSTR171. Hippocampal-Cortical Interactions I

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR171.11/QQ14

Topic: H.08. Learning and Memory

Support: CIHR

Title: Exploring the role of synchronous population events and preparatory behaviors in Spatial Navigation: insights from calcium imaging in freely behaving mice.

Authors: *J.-B. BOTT, L. PENAZZI, S. AL ACHKAR, S. WILLIAMS;
Psychiatry, McGill University, Montreal, Verdun, QC, Canada

Abstract: Spatial navigation relies on hippocampal synchrony to encode and recall key salient features of the environment. This includes an established neural activity pattern known as Sharp Wave Ripples (SWRs), which is instrumental in supporting memory formation. In parallel, Synchronous Population Events (SPEs), recorded using calcium imaging, may also be a relevant signature of spatial learning. Like SWRs, SPEs are predominantly recorded during awake immobility, including preparatory behaviors preceding active navigation. Our study aimed to analyze the spatiotemporal characteristics of SPE occurrence and their corresponding spatial information content. This was accomplished using calcium imaging of large CA1 neuronal population under conditions of successfully learning *versus* poor learning outcomes. We employed the spatial memory StarMaze task combined with freely behaving calcium imaging using head-mounted miniature microscopes (Miniscope). In this set-up, mice were trained to navigate in this apparatus to reach a rewarded target arm. As part of the experimental design to disrupt learning, we selectively eliminated access to distal cues during the preparatory behaviors prior to active navigation. Our findings revealed that SPEs were particularly prominent during the initial stages of learning, a phase where mice were becoming accustomed to the spatial context of the experiment. Successful learning was associated with SPEs encompassing information that spanned across the entire environment. In contrast, when preparatory behaviors were disrupted leading to impaired learning, SPEs displayed a bias toward abnormal replay of previous experiences. This underscores the crucial role of intact preparatory behaviors and SPEs in facilitating proper encoding and subsequent recall of spatial information.

Disclosures: J. Bott: None. **L. Penazzi:** None. **S. Al Achkar:** None. **S. Williams:** None.

Poster

PSTR171. Hippocampal-Cortical Interactions I

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR171.12/QQ16

Topic: H.08. Learning and Memory

Support: CIHR Foundation Program FDN-148478
NSERC Discovery Grant RGPIN-2020-06717

Title: Unraveling the distinct roles of hippocampal theta rhythms and spatiotemporal representations in memory functions using an all-optical approach

Authors: *G. ETTER[1], S. VAN DER VELDT[3], C.-A. MOSSER[2], S. WILLIAMS[1];
[1]Psychiatry, McGill Univ., Montreal, QC, Canada; [2]McGill Univ., Montréal, QC, Canada; [3]Univ. de Montréal, Montréal, QC, Canada

Abstract: The hippocampus is essential for episodic and working memory, and its neurons tend to fire in stable sequential patterns across sub-second and behavioral timescales. Whether this precise temporal coordination of activity is necessary for memory retrieval remains unclear. Within the medial septum, inhibitory neurons expressing parvalbumin provide extensive projections to the hippocampus and significantly contribute to the regulation of hippocampal theta (~8 Hz) oscillations. Using calcium imaging in freely behaving mice, we first find that hippocampal neurons encode a mixture of features, including location, time, and distance. We provide analytical accounts for how these representations emerge as a function of cognitive tasks and how they might be integrated into downstream regions, notably the lateral septum. We then dissociate the role of theta rhythms in spatiotemporal coding and memory using an all-optical interrogation and recording approach. Importantly, optogenetic interventions designed to abolish theta oscillations by scrambling septal inhibitory neurons modulate a portion of neurons in the hippocampus. This manipulation results in decreased episodic and working memory retrieval while leaving hippocampal spatiotemporal codes unaffected. This implies that while hippocampal theta rhythms are fundamental to memory functions, they can be dissociated from spatiotemporal representations, highlighting their distinct roles in memory processing.

Disclosures: G. Etter: None. **S. van der Veldt:** None. **C. Mosser:** None. **S. Williams:** None.

Poster

PSTR171. Hippocampal-Cortical Interactions I

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR171.13/QQ17

Topic: H.08. Learning and Memory

Support: CIHR

Title: Investigating OLM and VIP+ interneuron activity dynamics during anxiogenic and goal-directed tasks using calcium imaging in freely behaving mice

Authors: *S. AL-ACHKAR[1], L. PENAZZI[2], J.-B. BOTT[3], S. WILLIAMS[4];
[1]McGill Univ. Integrated Program in Neurosci., Montreal, QC, Canada; [2]McGill Univ., Montrea;, QC, Canada; [3]McGill University, Montreal, Verdun, QC, Canada; [4]McGill University, Douglas Res. Center,, Montreal, QC, Canada

Abstract: The hippocampal CA1 region is enriched with diverse GABAergic inhibitory interneurons, playing indispensable roles in the operational efficacy of neural networks. Notably, Oriens-Lacunosum Moleculare (OLM) cells, a subclass of somatostatin-positive hippocampal interneurons, can regulate information flow to CA1 pyramidal cells by gating input from the entorhinal cortex and CA3. Conversely, Vasoactive Intestinal Polypeptide-expressing (VIP+)

interneurons disinhibit CA1 pyramidal cells by inhibiting other classes of interneurons, including OLM cells. Recent evidence has shown that OLM cell activity is strongly modulated during locomotion, whereas VIP+ interneuron activity is intricately regulated in response to reward-associated behavior. However, a comprehensive understanding of the spatial-temporal dynamics of OLM and VIP+ cell activity during hippocampal-dependent and -independent tasks, particularly in freely behaving mice, remains elusive. This project aims to characterize OLM and VIP+ cell activity patterns during different anxiogenic and goal-directed behavioral tasks. Specifically, we aim to determine whether their activity evolves over the course of the task, and whether their activity is associated with different behavioral components such as exploration or familiarization. We performed calcium imaging of interneurons using head-mounted fluorescent miniature microscope (Miniscope) in freely moving mice. Chrna2-cre mice and VIP- cre mice were used to image OLM interneurons and VIP+ interneurons, respectively. We anticipate that the activity characterization data will reveal contrasting patterns of OLM and VIP+ activity. These opposing patterns are expected to mirror the differential inhibitory function enacted by OLM cells and the disinhibitory function performed by VIP+ cells. Unveiling these patterns could provide profound insights into the dynamic interplay of inhibitory networks within the hippocampal CA1 region.

Disclosures: **S. Al-Achkar:** None. **L. Penazzi:** None. **J. Bott:** None. **S. Williams:** None.

Poster

PSTR171. Hippocampal-Cortical Interactions I

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR171.14/QQ18

Topic: H.08. Learning and Memory

Support: NIH R01 AG070094
NIH R01 AA029700
FL DOH 20A09

Title: Neural dynamics in the HPC-PC Circuit underlying egocentric, allocentric and egocentric to allocentric reference frame transformation in a novel map to action transformation paradigm

Authors: *A. BREA GUERRERO[1], X. ZHOU[3], B. J. CLARK[5], W. WU[4], A. A. WILBER[2]; [1]Psychology, [2]Florida State Univ., Florida State Univ. Program In Neurosci., Tallahassee, FL; [4]Florida State Univ., [3]Florida State Univ., Tallahassee, FL; [5]Univ. of New Mexico, Univ. of New Mexico, Albuquerque, NM

Abstract: Navigating space and forming enduring memories are essential for animal survival. These abilities rely on interfacing between map-like or allocentric frameworks and a body-centered or egocentric reference frame. Fluid navigation involves converting between these reference frames such as recalling the layout of a city and then selecting the appropriate action (e.g., right turn). A parietal (PC)-anterior thalamic-hippocampal (HPC) network is considered

crucial for interfacing between frameworks. Our previous work explored the anterior thalamic nucleus-PC network's role in transforming allocentric place to egocentric action. In this study, we investigate the HPC-PC circuit. We aim to test the hypothesis that the HPC-PC circuit is essential for accessing map-like memory and generating the required action to reach a goal. To test this, we developed a novel behavioral task called the map-to-action transformation (MAT) task, involving rats navigating under three conditions: allocentric, MAT, and egocentric. Initially, rats are trained to navigate to a reward location on the perimeter of an 8-arm radial maze from one of seven start locations (arms) using distal cues. After reaching criterion, the animals proceed to the MAT paradigm. Rats are placed in a transparent box for 5 seconds at random start locations, followed by the introduction of an opaque box and curtains to obscure the distal cues. Rats must then remember the reward location and execute the appropriate egocentric action to reach it. In the final paradigm, the egocentric condition, the distal cues are concealed, while maintaining the egocentric relationship between the start and rewarded locations. The rats are expected to develop an egocentric strategy to locate the reward (e.g., second alley to the right), as they cannot rely on external cues. We found that rats can learn all three conditions of the MAT task. We also developed a decoding algorithm using spiking data from HPC and PC while the animal is either in the transparent box at the beginning of each trial or during the traversal from the start location to the center of the maze to predict the animal's movement from the center of the maze to the final location. We also tested the decoding model on spikes occurring during HPC sharp wave ripples. We aimed to determine the roles of the PC and HPC in each of the task phases. Furthermore, we analyzed the timing of the decoded information and put forth a hypothesis suggesting that, during the transformation task, HPC would first decode the allocentric goal location before subsequently decoding an action in PC. The decoded action code would represent the animal's action sequence to the goal location.

Disclosures: **A. Brea Guerrero:** None. **X. Zhou:** None. **B.J. Clark:** None. **W. Wu:** None. **A.A. Wilber:** None.

Poster

PSTR171. Hippocampal-Cortical Interactions I

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR171.15/QQ19

Topic: H.08. Learning and Memory

Support: FL DOH 20A09
R01 AG070094
NIA 1F31AG079619-01

Title: Resting After Learning Facilitated Memory Consolidation to Reverse Impairments in Spatial Reorientation in 3xTg-AD Mice

Authors: *S. C. MOSELEY, A. C. STIMMELL, J. MARQUEZ DIAZ, S. D. CUSHING, L. J. ALDAY, A. A. WILBER;
Psychology, Florida State Univ., Tallahassee, FL

Abstract: An essential component of productive memory consolidation and waste product clearance, including pathology associated with Alzheimer's disease (AD), is sleep (Tononi et al., 2014; Lucey 2020). Poor sleep is associated with increased amyloid beta (Aβ) and tau, poor cognition, and an increased risk of AD (Nebel et al 2018); while facilitation of sleep decreases Aβ and tau accumulation (Zhao et al., 2019). Sleep is also important for the consolidation of memories, including spatial memories (Ego-Stengel & Wilson, 2009; Jadhav et al, 2012; Maingret et al, 2016). Brain dynamics during rest are disrupted in 3xTg-AD female mice and contribute to impaired spatial navigation behavior (Benthem et al 2020). Thus, improving rest may offer a multifaceted approach to improving cognition and slow or halt disease progression in those at risk for AD; however, studies assessing sleep impacts on AD often fail to assess cognition.
Getting lost, particularly in new surroundings, is an early impairment in humans that will develop AD (Allison et al., 2016). Recent work including our own suggests that this early impairment, getting lost in new surroundings, could represent a failure to use distal cues to reorient in space (Stimmell et al., 2019). Thus, we set out to assess the impact of rest on impaired spatial reorientation previously observed in 6-month female 3xTg-AD mice. We randomly assigned 3xTg-AD mice to a rest (n = 7; 50 min pre- & post-task induced rest) or a non-rest group (n= 7; mice remained in the home cage pre- & post- task). Mice in both groups were compared to non-Tg age matched non-rest controls (n=7). Finally, to confirm that our sleep condition induced sleep we performed the same experiment in 3xTg-AD and control mice (n=6/group) implanted with recording electrodes in the hippocampus for recording local field potentials which were used to classify sleep states. Markers of pathology were also assessed in the parietal-hippocampal network where we previously showed pTau positive cell density predicts spatial reorientation ability (pTau, 6e10, M78, M22, as in Stimell et al., 2020) but here automatically counted with Zeiss ZEN (blue edition) 3.7 software. We found that 6-month female 3xTg-AD sleep mice (both with and without a recording array) were not impaired at spatial reorientation, while 6-month female 3xTg-AD no sleep mice were impaired at spatial reorientation learning, a replication of Stimmell et al., 2019. This recovered behavior persisted despite no change in the density of pathology positive cells. Thus, improving sleep in early stages of AD pathology offers a promising approach for facilitating memory consolidation to improving cognition.

Disclosures: S.C. Moseley: None. **A.C. Stimmell:** None. **J. Marquez Diaz:** None. **S.D. Cushing:** None. **L.J. Alday:** None. **A.A. Wilber:** None.

Poster

PSTR171. Hippocampal-Cortical Interactions I

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR171.16/QQ20

Topic: H.08. Learning and Memory

Support: NIA R00 AG049090
FL DOH 20A09
F32MH099682
R01 AG070094
NIAAA R01 AA029700

Title: A Hippocampal-parietal Network for Map to Action Transformation

Authors: *Y. ZHENG[1], X. ZHOU[2], S. MOSELEY[3], B. J. CLARK[4], W. WU[2], A. A. WILBER[3]; [1]Florida State Univ. Program In Neurosci., Tallahassee, FL; [2]Dept. of Statistics, [3]Psychology, Florida State Univ., Tallahassee, FL; [4]Univ. of New Mexico, Univ. of New Mexico, Albuquerque, NM

Abstract: Movement through space and establishing memories based on such experiences is essential for survival. This ability is thought to require both an allocentric (map-like) and body-centered 'maps' of our surroundings which must be coordinated in a fluid manner during navigation. The encoding observed in hippocampus (HPC) and parietal cortex (PC) has led to the notion that this circuit operates as a part of a coordinate transformation network; for example, transforming a remembered allocentric representation into the appropriate action. Our previous work dissected an anterior thalamic nucleus-PC circuit within this network that appears critical for the *transformation* between allocentric place and egocentric action. Here we examine the HPC-PC circuit to test the hypothesis that allocentric information is conveyed by the HPC to the PC for conversion to egocentric action. While HPC neurons are typically modulated by allocentric locations, PC neurons are modulated by multiple reference frames including actions (e.g., right turn). This hypothesis was tested using a *complex spatial sequence task* where rats learn to navigate to unmarked locations fixed in space in a specific sequence. Landmarks are distributed around the room for spatial orientation. We use a sequence (**1-2-3**-4-**1-2-3**-5-) that has a repeating path segment (**1-2-3**) followed by one of two distinct actions. Specifically, the rat learns in context 5-**1-2-3**, to go to 4 for reward, while in context 4-**1-2-3** the rat must go to 5. Thus, navigation to zone 4 or 5 requires remembering the allocentric context and converting to the appropriate action. This emulates the spatial memory problem one encounters when driving through an intersection and remembering the appropriate action given the current route and goals (e.g., turn left to a bank versus right to home). Thus, the rat must coordinate between remembered allocentric context and egocentric action. We found that HPC single units encode the allocentric context during the **1-2** segment while PC ensembles encode action information for the upcoming action during the **2-3** segment. Surprisingly, the allocentric context signal is also apparent in PC close in time to the emergence of the HPC allocentric context signal. Finally, the time lags between spatial and action codes appear to be fixed at ~2s, a surprisingly long delay for the number of synapses theoretically involved in this transformation. Thus, we provide evidence that the HPC-PC circuit is a key component of a coordinate transformation network for interfacing between map-like and person-centered reference frames.

Disclosures: Y. Zheng: None. **X. Zhou:** None. **S. Moseley:** None. **B.J. Clark:** None. **W. Wu:** None. **A.A. Wilber:** None.

Poster

PSTR171. Hippocampal-Cortical Interactions I

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR171.17/QQ21

Topic: H.08. Learning and Memory

Support: NIA R00 AG049090
FL DOH 20A09
R01 AG070094
NIA F31AG079619-01

Title: Rescuing impaired cortical-hippocampal interactions during sleep with hippocampal 40Hz stimulation in 3xTg-AD/PVcre mice

Authors: *S. CUSHING[1], S. C. MOSELEY[2], A. STIMMELL[1], A. A. WILBER[3];
[2]Dept. of Psychology, [1]Florida State Univ., Tallahassee, FL; [3]Psychology, Florida State Univ. Program In Neurosci., Tallahassee, FL

Abstract: In preclinical Alzheimer's disease (AD), spatial learning and memory is impaired (Allison et al., 2016). We reported similar impairments in 6-month 3xTg-AD female mice on a virtual *spatial-reorientation-task* (VM) that requires learning to use landmarks to navigate (Cushing et al., 2020). Memory replay during sleep is critical for learning related plasticity (Ego-Stengel & Wilson, 2009; Jadhav et al, 2012; Maingret et al, 2016), and hippocampal (HPC)-cortical dysfunction is a potential mechanism for memory impairments in AD (Gennaro et al, 2017; Khan et al, 2014). We previously found deficits in HPC-parietal cortex (PC) coordination during sleep coinciding with impairments on the VM (Cushing et al, 2020). Various forms of 40Hz stimulation have been shown to clear AD pathology in mice (Iaccarino et al, 2016 - optogenetic & light; Martorell et al, 2019 – combined light/sound), and improve functional connectivity in preclinical AD patients (He et al, 2021 combined light/sound), though a recent paper showed lack of improvement from light only 40Hz (Soula et al, 2023). Thus, we assessed HPC-PC coordination in 3xTg-AD/PV^{cre} mice learning the VM. We implanted a 16-tetrode recording array targeting PC and HPC and an optical fiber targeting HPC. Daily recording sessions of rest-task-rest began as mice learned the VM, followed by HPC optogenetic stimulation, either 40Hz or SHAM. We assessed sleep quality metrics, delta waves (DW) in PC, and HPC markers of memory replay (SWRs) during slow wave sleep. Only 40Hz stimulation increased 40Hz power in HPC and PC. In SHAM stimulated mice SWR-DW cross-correlations were reduced, similar to 3xTg-AD mice (Cushing et al, 2020). In 40Hz stimulated mice, this phase-locking was rescued. Furthermore, the 40Hz stimulation restored performance on the VM compared to SHAM mice. However, this rescued HPC-PC coupling no longer predicted performance as in NonTg animals (Cushing et al, 2020). Instead, DWs independently predicted performance in 40Hz mice only. We also found reduction of amyloid beta (Aβ) in PC and ventral CA1, but no reduction in dorsal CA1, synaptically closer to stimulation. There was also no difference in astrocyte or microglia density, and neither glial cell density correlated with Aβ or tau load. Thus, 40Hz stimulation of HPC rescued learning and memory related functional interactions in the HPC-PC network during sleep and as a consequence rescued impairments in

spatial navigation, despite a decoupling between HPC-PC coupling and learning and memory. Furthermore, the observed effects seems to be mediated by direct 40Hz impacts on brain dynamics, and not 40Hz stimulation effects on glia, Aβ, or tau contrasting previous reports.

Disclosures: **S. Cushing:** None. **S.C. Moseley:** None. **A. Stimmell:** None. **A.A. Wilber:** None.

Poster

PSTR171. Hippocampal-Cortical Interactions I

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR171.18/QQ22

Topic: H.08. Learning and Memory

Support: National Institutes on Aging R01AG062762

Title: Alterations in cortico-hippocampal dynamics in a hyperglycemic mouse model for Alzheimer's disease

Authors: *E. FLORES[1,4], A. A. ORTIZ[4], R. A. WIRT[1], J. V. TRINIDAD[5], D. C. BRAGANZA[5], D. A. MINOR[5], M. PIZIO[5], J. W. KINNEY[2], J. M. HYMAN[3];
[1]Univ. of Nevada Las Vegas, LAS VEGAS, NV; [2]Brain Hlth., [3]Psychology, Univ. of Nevada Las Vegas, Las Vegas, NV; [4]Interdisciplinary neuroscience, [5]Univ. of Nevada, Las Vegas, Las Vegas, NV

Abstract: Alzheimer's disease (AD) is characterized by the progressive loss of memory and cognitive function, driven by notable pathology consisting of hyperphosphorylation of tau, aggregation of amyloid beta, and an increased inflammatory response. Closely related, diabetes mellitus (DM) is a known risk factor for the development of AD with a staggering 65% increased chance of developing AD. DM is characterized by hyperglycemia and patients with DM also have neurological impairments, including deterioration of working memory, diminished information processing, deficits in attention, concentration, and executive functions. Both AD and DM lead to an increased inflammatory response in the brain with the activation of microglia, which could potentially impact neuronal activity in these areas. Our previous work has shown alterations in spatial working memory in a rat model of chronic hyperglycemia that were similar to changes observed in mouse models of AD pathology. We expand on this work by examining network activity in mice, to allow for a more direct comparison to AD mouse models, and also by moving mice between several glycemic states we could better understand how blood glucose levels themselves might be impacting spatial cognition-related signals. We hypothesized that the activated immune response was responsible for the learning deficits and altered theta activity and that this was the primary mechanism by which DM leads to cognitive deficits, similar to AD. To examine these changes we administered low dose, intermittent streptozotocin to mice (N=8), then after reaching hyperglycemic states we administered phloridzin (PZ) to reach normoglycemic levels. During each of these glycemic states we recorded from the hippocampus and anterior cingulate cortex (ACC). Our results show alterations in hippocampal power in delta

(F(3,21)=6.89,p=0.0021), theta (F(3,21)=3.85,p=0.0243), and beta (F(3,21)=3.33,p=0.039) range. We also observed differences in theta/delta ratio (F(3,21)=6.49, p=0.0028) and significant differences in hippocampal-ACC theta coherence (F(3,41)=6.61,p=0.001) between baseline and after PZ treatment day. Our results suggest that treating hyperglycemia with PZ does not reverse the effects observed during hyperglycemic states, but may lead to some unique oscillatory effects that differ from both baseline and hyperglycemic states. Overall, we found that oscillatory changes in hippocampal activity that accompany chronic hyperglycemia were not remedied by restoring blood glucose levels and achieving normoglycemia, suggesting that such changes were due to other factors as opposed to blood glucose levels.

Disclosures: **E. Flores:** None. **A.A. Ortiz:** None. **R.A. Wirt:** None. **J.V. Trinidad:** None. **D.C. Braganza:** None. **D.A. Minor:** None. **M. Pizio:** None. **J.W. Kinney:** None. **J.M. Hyman:** None.

Poster

PSTR171. Hippocampal-Cortical Interactions I

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR171.19/QQ23

Topic: H.08. Learning and Memory

Support: NIGMS P20GM109025

Title: From beneficial to detrimental: Acute and chronic neuroinflammation uniquely influence oscillations in the hippocampus and anterior cingulate cortex

Authors: *L. A. CREW[1], A. A. ORTIZ[1], D. C. BRAGANZA[1], J. V. TRINIDAD[1], M. PIZIO[1], J. W. KINNEY[1], J. M. HYMAN[2];
[2]Psychology, [1]Univ. of Nevada Las Vegas, Las Vegas, NV

Abstract: Neuroinflammation is a distinctive pathological feature in numerous neurodegenerative diseases, including Alzheimer's disease; however, our understanding of whether and how neuroinflammation influences the symptoms of these conditions remains limited. The inflammatory state can often lead to brain degeneration, loss of neurogenesis, and localized atrophy in the hippocampus and anterior cingulate cortex (ACC). These areas are known to be involved in learning and can operate together to maintain and manipulate information used to guide future responses. Acute neuroinflammation is beneficial, even essential, to combat foreign entities in the brain and promote healing, but as inflammation persists and becomes chronic it can also become detrimental. What changes occur during this transition? By drawing direct comparisons between acute and chronic neuroinflammation, we aimed to pinpoint alterations in neural activity in the hippocampus and ACC. Our goal was to better understand the transition from acute to chronic stages and identify potential factors that could explain the symptoms associated with chronic neuroinflammation. We recorded activity from the hippocampus and ACC in behaving mice to characterize cognition-linked oscillatory

activity in combination with acute and chronic protocols of polyinosinic:polycytidylic acid (poly I:C) to provoke inflammation. Using a within subject protocol, we found unique electrophysiological profiles for both acute and chronic poly I:C mice. Both acute and chronic neuroinflammation resulted in deviations from baseline levels across multiple oscillatory bands, including increased beta power in the hippocampus, possibly suggesting heightened sensory responsiveness. As for theta oscillations, known for their strong association with memory processes in the hippocampus, we observed an increase in power exclusively in the chronic condition. Interestingly, alterations in both theta and beta oscillations have been observed in human patients with Alzheimer's and in various animal models simulating Alzheimer's pathology. Multiple cognitive processes, including working memory and cognitive flexibility, are dependent on theta interactions between the hippocampus and ACC. Our research revealed that acute neuroinflammation significantly amplified theta coherence between the hippocampus and ACC, while chronic neuroinflammation maintained baseline coherence levels.This suggests some form of neural adaptation was taking place during the transition from acute to chronic stages, which could provide a potential electrophysiological biomarker for this poorly understood transition.

Disclosures: L.A. Crew: None. **A.A. Ortiz:** None. **D.C. Braganza:** None. **J.V. Trinidad:** None. **M. Pizio:** None. **J.W. Kinney:** None. **J.M. Hyman:** None.

Poster

PSTR171. Hippocampal-Cortical Interactions I

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR171.20/QQ24

Topic: H.08. Learning and Memory

Title: Temporal integration of experiences in ACC ensemble activity

Authors: *T. SOLUOKU[1], J. M. HYMAN[2], R. A. WIRT[3], R. M. RICCI[4,5];
[1]Univ. of Nevada, Las Vegas Neurosci. Grad. Program, Las Vegas, NV; [2]Dept. of Psychology, Univ. of Nevada, Las Vegas, Las Vegas, NV; [3]Dept. of Psychology, Univ. of Nevada, Las Vegas,, Las Vegas, NV; [4]Kirk Kerkorian Sch. of Med., Univ. of Nevada, Las Vegas, NV; [5]Dept. of Med., Kirk Kerkorian Sch. of Med., Las Vegas, NV

Abstract: Anterior cingulate cortex (ACC) neurons play a critical role in processing internal and external stimuli experienced over time and generating responses based on relating these two streams of information. However, there is still a limited understanding of how ACC neurons integrate temporal information on behavioral timescales to facilitate rule learning, flexible behavior, and the differentiation of working memory trials to prevent interference. To examine temporal information coding in the ACC, we recorded large ensembles of neurons while rats completed a decision-free three-arm bandit task (n=5). We found that behavior switched soon after a within-session reward probability reversal. However, when we examined ensemble activity over trials, we found that neither behavioral preference nor reward probability

information was apparent in low-dimensional representations of ACC ensemble activity. Instead, the signals were predominantly influenced by a slow drifting signal that corresponded to the pace of behavioral performance, explaining approximately 9.5% of the overall ensemble variance. Remarkably, these signals exhibited very high consistency between ensembles, and using neural decoding techniques, we could accurately predict which trial number a rat was completing using a model based upon a different animal. Such effects were apparent in the background activity outside of trial behaviors, and we found that ramping firing rates over long periods of time (tens of minutes) were driving the low dimensional ensemble representations. We show that a pool of different ramp lengths was present in ACC ensembles and that combining different short duration ramping neurons together can create an ensemble that tracks longer durations. Lastly, we show that these temporal signals were multiplexed along with spatial and outcome information, while also being found in neurons without clear behavioral correlates. Collectively, these results provide insights into an ensemble-level temporal signal in the ACC that directly reflects the accumulation of experiences over prolonged periods.

Disclosures: **T. Soluoku:** None. **J.M. Hyman:** None. **R.A. Wirt:** None. **R.M. Ricci:** None.

Poster

PSTR171. Hippocampal-Cortical Interactions I

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR171.21/QQ25

Topic: H.08. Learning and Memory

Support: National Institute on Aging R01AG062762

Title: <Fragmented spatial maps and compromised reward coding in the cortico-hippocampal loop in a hyperglycemia risk factor model of Alzheimer's disease.>

Authors: ***G. BHASIN**[1], L. A. CREW[2], A. A. ORTIZ[2], R. A. WIRT[2], E. FLORES[2], J. W. KINNEY[3], J. M. HYMAN[4];
[1]Univ. of Nevada, Las Vegas, las vegas, NV; [3]Brain Hlth., [4]Psychology, [2]Univ. of Nevada Las Vegas, Las Vegas, NV

Abstract: Alzheimer's disease (AD) is characterized by amyloid beta plaques, hyperphosphorylated tau tangles and neuroinflammation, leading to progressive cognitive decline and inability to carry out daily functions. Neuroinflammation is also linked to Type 2 diabetes (DM2), characterized by hyperglycemia, and affects spatial and cognitive functions in the cortico-hippocampal loop. After going through an intermittent, low dose streptozotocin (STZ) protocol, which leads to lasting hyperglycemia, single unit activity was recorded from the anterior cingulate cortex (ACC) and hippocampus (CA1), as hyperglycemic (n=5) and control (n=3) rats performed a variable length delayed alternation task on a figure 8-shaped T-maze. In both areas we found a higher percentage of place cells and spatial information in STZ versus controls. Spatial information in hippocampal (HC) neurons was higher for locations leading up to

the reward zone in the hyperglycemic (STZ) group, while controls animals had more post reward location information. Ensemble analysis focused on population decoding of left and right (L/R) trials at different locations in the maze. Hippocampal STZ ensembles had larger state space separation between L/R trials over the entire maze, ensemble decoding accuracy was the highest at reward, and unlike in controls, L/R separation continued through the return arms of the maze. We hypothesize that this could indicate the use of two separate, fragmented spatial maps to traverse the maze (one for left and one for right trials) by STZ animals, as opposed to a singular map in controls. In contrast, the only difference between ACC ensemble groups in state separation between L/R trials was at the reward location, with better separation in controls. Decoding accuracy analysis revealed the strongest coding at the reward location in controls compared to STZ, indicative of reward overrepresentation in ACC control ensembles. Lastly, ACC hippocampal theta phase-locked cells had stronger reward coding than non-theta cells, indicating reward overrepresentation in ACC ensembles was driven by theta phase-locked cells, and there was specific deficit in reward related information in ACC hippocampal theta phase-locked cells. Our results indicate altered reward representation in hyperglycemic animals, whereby HC single units strongly coded for reward, but ensembles did not, while ACC control ensembles coded for reward overrepresentation, but the single units did not. Consequently, hyperglycemia affects ACC and HC spatial coding in vastly different ways at the single unit level and at ensemble level, bringing varied influence at both levels of information processing in the brain.

Disclosures: G. bhasin: None. **L.A. Crew:** None. **A.A. Ortiz:** None. **R.A. Wirt:** None. **E. Flores:** None. **J.W. KInney:** None. **J.M. Hyman:** None.

Poster

PSTR172. Learning and Memory: Physiology

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR172.01/QQ26

Topic: H.08. Learning and Memory

Title: Fear Conditioning Associated Neuronal Activation in the Cerebellum

Authors: *A. ABDULLA[1], S.-Q. LIU[2];
[2]Cell Biol. and Anat., [1]LSU Hlth. Sci. Ctr., New Orleans, LA

Abstract: The cerebellum is critically involved in cognitive functions and the formation of emotional memories, and its dysfunction leads to neurological disorders such as autism spectrum disorder, schizophrenia, addiction, and PTSD. Fear conditioning is an associative learning paradigm that is used to identify the molecular and circuitry mechanisms of emotional memory disorders. This learning paradigm activates sparse ensembles of neurons and subsequent strengthening of the connections among these neurons make them an “engram” that is believed to be the neural representation of the memory. Indeed, fear memory engrams have been identified in the amygdala, hippocampus and cortex. The cerebellum forms extensive

connections with the limbic system, and inactivation of cerebellar activity disrupts the consolidation of associative fear memory. Our recent studies show the activity of cerebellar molecular layer interneurons (MLIs) and neural plasticity of these neurons are required for memory consolidation, suggesting that the cerebellar cortex could be a site where the fear memory engram resides. We used targeted recombination in active populations (TRAP) to indelibly label active neurons with fluorescent protein during fear conditioning and found that fear conditioning increases neuronal activation in the cerebellar vermis. We measured the activation of MLIs and Purkinje cells in cerebellar lobules I-X and found increased MLI activation per unitary area and volume in lobules V/VI when compared to both unpaired and naïve controls. This results suggest that cerebellar MLIs are activated during associative fear learning and memory consolidation.

Disclosures: **A. Abdulla:** None. **S. Liu:** None.

Poster

PSTR172. Learning and Memory: Physiology

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR172.02/QQ27

Topic: H.08. Learning and Memory

Support: NRF-2022R1A2C1012351
NRF-2019R1F1A1063005

Title: Effects of diverse stress on the learning and memory functions in C57BL/6 mice.

Authors: *C. LEE, J.-H. JANG;
Dept. of Pharmacol., Sch. of Medicine, Keimyung Univ., Daegu, Korea, Republic of

Abstract: Stress is regarded as one of the critical risk factors for neurodegeneration leading to learning and memory impairment. However, recently a possibility of mild stress to potentiate cognitive functions has been reported by several researchers, whereas its underlying molecular mechanisms are not clearly verified. In this study we have investigated the effect of stress on the learning and memory functions in C57BL/6 mice by conducting a series of behavior tests and molecular analyses. Unpredictable chronic stress (UCS) for 28 days in C57BL/6 mice induced depression-like symptoms and increased the susceptibility to cognitive dysfunction as assessed by fear conditioning test. Conversely, predictable chronic mild stress (PCMS) improved mean escape latency, the time taken to find the platform during training trials in Morris water-maze test. To further elucidate the molecular mechanisms, we have examined the molecules related with oxidative stress and neuroinflammation. UCS caused oxidative damages and inflammatory responses in the brain, which were ameliorated by PCMS through up-regulation of antioxidant enzymes via activating NF-E2-related factor. Stress can work with or against memory depending on the type of stress and controllable moderate level of stress may have a potential beneficial effect.

Disclosures: C. Lee: None. **J. Jang:** None.

Poster

PSTR172. Learning and Memory: Physiology

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR172.03/QQ28

Topic: H.08. Learning and Memory

Title: Role of binaural beats in sleep and cognitive functions in young healthy adults.

Authors: *H. KAUSER;
Physiol., Vardhman Mahavir Med. Col. and Safdarjung Hosp., Delhi, India

Abstract: Background: Sleep and cognitive dysfunction is most common problem in health and disease. Various non-invasive techniques i.e., transcranial direct current stimulation and transcranial magnetic stimulation are used to ameliorate these deficits. Most recently, stimulation with binaural beats has been emerged as best tool to improve various aspects of sleep and cognition. In the present study, we planned to evaluate the efficacy of binaural beats stimulation on various functions of sleep and cognition in young healthy adults.**Methods:** The subjects were screened for their sleep by PSQI. Their cognitive abilities were assessed by CBS neuropsychological battery and P300 recordings. The sleep disturbed individuals were undergone auditory stimulation for 30 minutes just before sleep each day for four weeks. The outcome measures were obtained at baseline, after 2 weeks and 4 weeks of intervention. **Results:** We found a significant decrease in sleep and cognition in 60% of healthy individuals. A significant improvement in sleep and cognition was reflected on various measures of PSQI and CNB with 4 weeks of auditory stimulation. Moreover, P300 measures, i.e., amplitude and latency both increased significantly with the same protocol of auditory stimulation. **Conclusion:** These results indicate that binaural beats stimulation during sleep has a potential to improve sleep quality and cognitive functions.**Ethics statement:** The current study was approved in compliance with the guidelines set out by the Institutional Ethics Committee.**Acknowledgement:** The authors would like to acknowledge the university students who participated in the study.

Disclosures: H. Kauser: None.

Poster

PSTR172. Learning and Memory: Physiology

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR172.04/RR1

Topic: H.08. Learning and Memory

Title: Fasting of youth makes lifelong improvement of cognitive function in Drosophila

Authors: *M.-C. HSU;
Basic Med. Sci., Natl. Cheng Kung Univ. Col. of Med., tainan, Taiwan

Abstract: The brain plays a crucial role in the formation of memory in animals, enabling them to adapt to a changing world and ensuring their survival. Dopaminergic neurons (DANs) are particularly significant in the brains of animals, as they regulate various signals related to appetitive behavior, energy homeostasis, reward-based learning, and aversive learning (Aso et al., 2014; Han et al., 2016; Ter Horst et al., 2018). In Drosophila, two clusters of DANs, namely PAM and PPL1, function as nutrition sensors, integrating information about hunger and satiety into the mushroom body (MB), which serves as the learning center in flies (Huetteroth et al., 2015; Tsao et al., 2018). However, the mechanisms underlying memory formation in different situations and the regulation of memory in response to dynamic environments remain unknown. In our study, we investigated the effects of 18 hours of food deprivation (18-fd) on memory formation. We discovered that this period of food deprivation enhanced short-term memory (STM) in young flies but not in older flies. Interestingly, the improvement in STM observed in young flies was maintained as they aged. Our findings also revealed the involvement of two specific circuits, namely PAM-γ3 and PPL1-γ2α′1, following 18-fd. This suggests that internal states can shape brain circuits by incorporating new components into the existing circuitry, which can then be maintained throughout the aging process.

Disclosures: M. Hsu: None.

Poster

PSTR172. Learning and Memory: Physiology

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR172.05/RR2

Topic: H.08. Learning and Memory

Support: ERC Starting Grant AXPLAST
SNF Professorship (J.G.)
AMBIZIONE Fellowship (J.G.)

Title: Axon initial segment dynamics during associative fear learning.

Authors: D. A. GANEA[1,2], C. M. BENOIT[1,2], ***R. PARICIO MONTESINOS**[1], C. THOME[3,4], A. SATTIN[5], S. M. INNOCENTI[2], S. KRABBE[1], A. LÜTHI[6,2], T. FELLIN[5], M. ENGELHARDT[3], J. GRÜNDEMANN[1,2];
[1]German Ctr. for Neurodegenerative Dis. (DZNE), Bonn, Germany; [2]Univ. of Basel, Basel, Switzerland; [3]Johannes Kepler Univ., Linz, Austria; [4]Inst. for Stem Cell Biol. and Regenerative

Med., Stanford Univ., Stanford, CA; [5]Inst. Italiano di Tecnologia, Genova, Italy; [6]Friedrich Miescher Inst., Basel, Switzerland

Abstract: The axon initial segment (AIS) is the site of action potential initiation and plays a crucial role in the generation of neuronal activity and the maintenance of network function during sensory processing and learning. While previous *ex vivo* studies identified the AIS as a site of homeostatic plasticity, the occurrence of structural changes of AIS *in vivo* and their implication in learning remains unknown. By performing *in vivo* longitudinal two-photon imaging of live-stained AIS in the mouse medial prefrontal cortex, we reveal dynamic AIS length remodelling during associative fear learning and extinction. Notably, we observed distinct bidirectional AIS plasticity mechanisms that may balance the excitability of neuronal subnetworks with distinct functions in memory formation and extinction, which could ultimately influence behavior and memory recall. Our findings suggest that axon initial segment dynamics are not only crucial for homeostatic adaptation but also act as a hallmark of memory formation.

Disclosures: D.A. Ganea: None. **C.M. Benoit:** None. **R. Paricio Montesinos:** None. **C. Thome:** None. **A. Sattin:** None. **S.M. Innocenti:** None. **S. Krabbe:** None. **A. Lüthi:** None. **T. Fellin:** None. **M. Engelhardt:** None. **J. Gründemann:** None.

Poster

PSTR172. Learning and Memory: Physiology

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR172.06/RR3

Topic: H.08. Learning and Memory

Support: NIH T32ES007141-38
JHU SURPASS GRANT

Title: Modulation of Network Dynamics in hiPSC Derived Brian Organoids Towards Studying Synaptic Plasticity in Vitro

Authors: *D.-M. ALAM EL DIN[1,2], L. MÖNKEMÖLLER[2], L. DONG[1,2], J. SCHENKMAN[3], A. LOEFFLER[4], B. KAGAN[4], T. HARTUNG[1,2], L. SMIRNOVA[1,2];
[1]Johns Hopkins Univ., Baltimore, MD; [2]Ctr. for Alternatives to Animal Testing, Baltimore, MD; [3]Dept. of Electrical and Computer Engin., Princeton Univ., Princeton, NJ; [4]Cortical Labs Pty Ltd, Melbourne, Australia

Abstract: Cognitive endpoints are used in human and animal studies to determine if a chemical will cause neurodevelopmental toxicity in drug screening. Currently, there are no *in vitro* assays that can be used to assess cognition, therefore, to determine the effect a chemical has on learning and memory, animal models are used. Recent advances in cell culture including Microphysiological Systems (MPS) allow more physiologically relevant modeling of the cellular processes in vitro and bring the *in vitro* models closer to *in vivo*. Our lab established a highly standardized and reproducible hiPSC derived brain organoid MPS consisting of all neural cell

types found in the central nervous system, myelinated axons, and spontaneous local field potentials. Using this MPS we aim to assess the extent of network modulation in brain organoids to determine the effect a chemical has on synaptic plasticity so this platform can be used for high throughput drug screening. To research this, we have been differentiating brain organoids for up to 12 weeks and characterizing spontaneous electrical activity using live calcium imaging and HD MEAs. In addition, we characterized the molecular components of early synaptogenesis, including immediate early genes (IEGs) and miRNAs over time in conjunction with chemical exposure by immunohistochemistry and RT-PCR. Lastly, a theta-burst stimulation LTP induction protocol was used to modulate hiPSC brain organoid activity electrically. Organoids showed spontaneous electrical activity using both HD MEAs and calcium imaging and overtime showed evidence of highly interconnected network formation within and between organoids. Additionally, we saw time-point specific expression of IEGs over development, which are known to be involved in synaptic plasticity. Our results also show that receptor agonists and antagonists impact IEG expression and electrical activity. Finally, theta-burst stimulation seemed to change overall network firing and activity suggesting changes in synaptic plasticity. In the future, we plan to build upon this model to study how long-term potentiation is affected by chemical and electrical exposures to develop a model to perform high throughput chemical screening to assess functional changes in synaptic plasticity. An in vitro cognition assay developed for hiPSC brain organoid MPS can fill the gap between human and animal data while increasing throughput for drug screening to test for the effect of chemicals on neurodevelopment and neurodegeneration.

Disclosures: **D. Alam El Din:** None. **L. Mönkemöller:** None. **L. Dong:** None. **J. Schenkman:** None. **A. Loeffler:** A. Employment/Salary (full or part-time); Cortical Labs. **B. Kagan:** A. Employment/Salary (full or part-time); Cortical Labs. **T. Hartung:** None. **L. Smirnova:** None.

Poster

PSTR172. Learning and Memory: Physiology

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR172.07/RR4

Topic: H.08. Learning and Memory

Support: CONACYT 855559

Title: Prenatal cafeteria diet primes anxiety-like behavior associated to defects in volume and diffusion in the fimbria-fornix of mice offspring

Authors: *G. CRUZ[1], L. TRUJILLO-VILLARREAL[1], D. ANGELES-VALDEZ[2], L. CONCHA[3], E. A. GARZA-VILLARREAL[4], A. CAMACHO[1];
[1]Univ. Autónoma De Nuevo León, Monterrey, Mexico; [2]Inst. de Neurobiología. Univ. Nacional Autónoma de México (UNAM), Juriquilla, Mexico; [3]Univ. Nacional Autonoma De Mexico, Monterrey, Mexico; [4]Inst. De Neurobiología, UNAM, Monterrey, Mexico

Abstract: Exposure to external stimuli during prenatal period provides a time-dependent modulation of physiological outcomes after birth, known as fetal programming (Gawlińska et al., 2021). Prenatal exposure to high-energy diets primes cognitive impairment, affecting learning and memory processes in the offspring after life (Liu et al., 2021). Some reports documented that offspring exposure to high-energy diets developed hippocampal-dependent behavioral abnormalities (Peleg-Raibstein, 2021). While still under investigation, a positive energy balance such as happens during obesity or prenatal exposure to high-energy diets is also able to promote structural brain abnormalities in infants and in adult individuals (Verstynen et al., 2012; Ward et al., 2005). However, the contribution of prenatal exposure to high-energy diets on structural brain conformational and activity coding for aberrant behavior after birth has not been documented. We used female C57BL6 mice (n=10) exposed to a high-energy diet (Cafeteria diet (CAF)) or Chow diet for 9 weeks (before, during, and after pregnancy) to characterize their effect on brain structural organization and learning and memory performance in the offspring at two-month-old (n=17). Memory and learning performance were evaluated using the Y-maze test including forced and spontaneous alternation, novel object recognition (NORT), open field, and Barnes maze tests. Imaging was performed in 7 T Bruker scanner. We then acquired 2 sequences: T1-weighted and Diffusion-weighted images (DWI). We found no alterations in the short- or long-time spatial memory performance in male offspring prenatally exposed to CAF diet when compared to the control, but they increased time spent in the edges resembling anxiety-like behavior. By using deformation-based morphometry and diffusion tensor imaging analysis we found that male offspring exposed to CAF diet showed increased volume in the primary somatosensory cortex and a reduced volume of fimbria-fornix, which correlate with alterations in its white matter integrity. We performed a multiple linear regression to predict the association between diet (CAF vs Chow), volume changes and dMRI metrics in the fimbria-fornix to behavioral outcomes in the offspring. Accordingly, dMRI metrics and fimbria-fornix volume found in the offspring prenatally exposed to CAF diet significantly predicts higher time spent in the edges during the open field tests, suggesting a higher anxiety score. We propose that structural defects of the fimbria-fornix might be a major predictor to anxiety in subjects prenatally exposed to CAF diet.

Disclosures: **G. Cruz:** None. **L. Trujillo-Villarreal:** None. **D. Angeles-Valdez:** None. **L. Concha:** None. **E.A. Garza-Villarreal:** None. **A. Camacho:** None.

Poster

PSTR172. Learning and Memory: Physiology

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR172.08/RR5

Topic: H.08. Learning and Memory

Title: Association between gut microorganisms and cognitive impairment inHIV and TB patients.

Authors: *L. E. AZUARA ALVAREZ[1], N. A. CASTILLO MARTINEZ[2], M. L. GARCIA GOMAR[3], A. NEGRETE CORTEZ[1], J. ALVELAIS PALACIOS[1], J. R. CHAVEZ MENDEZ[1]; [2]microbiology, [1]Univ. Autónoma de Baja California, Tijuana, Mexico; [3]Univ. Autónoma de Baja California, tijuana, Mexico

Abstract: HIV has shown tropism for the central nervous system, affecting patients progressively and significantly from early stages of infection. Some of the most important conditions reported in these patients are memory, learning and attention. One of the most frequent comorbidities of HIV is tuberculosis (HIV/TB), being 20 to 30 times more frequent in Tijuana, Mexico. Dysbiosis of the gastrointestinal microbiota has been shown to have a great relevance in the neurocognitive function of different pathologies and HIV/TB is no exception. Previously unpublished results from our research group show neurocognitive impairment in 84 patients, tested by Hopkins Learning Verbal Test-Revised (HLVT-R) and Paced Auditory Serial Addition Test (PASAT). For HLVT-R Total and Delayed Recall Scores, PLWHA showed a mean percentile of 40 (SD=29.7) and 30 (SD=32.7) using Mexican norms. We propose that by incorporating probiotics in the diet of HIV/TB patients we will observe a reduction in dysbiosis and this could improve the neurocognitive state and quality of life. Two groups of HIV/TB patients will be randomized in CTL=without probiotic intake and PB=with probiotic intake for 14 days. HLVT-R and PASAT and EEG tests will be performed to determine alterations in learning, memory and attention and neuronal activation pattern, stool samples will be taken for sequencing (16s) and culture (Bacteroidetes/Firmicutes) in addition to blood for CD4 quantification, liver enzymes, hematic biometry and blood chemistry to obtain a biochemical profile. Finally, a comparison will be made between the two groups in the variables described above. We expect to observe a significant difference in neurocognitive and biochemical tests between the PB and CTL group mediated by probiotic intake in the PB group.

Disclosures: L.E. Azuara Alvarez: None. **N.A. Castillo Martinez:** None. **M.L. Garcia Gomar:** None. **A. Negrete Cortez:** None. **J. Alvelais Palacios:** None. **J.R. Chavez Mendez:** None.

Poster

PSTR172. Learning and Memory: Physiology

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR172.09/RR6

Topic: H.08. Learning and Memory

Support: CIHR Project Grant

Title: Parvalbumin Neuron Calcium Dynamics During a Touchscreen Test of Spatial Working Memory

Authors: *S. HAMIDULLAH[1,2], T. D. DEXTER[1,2], D. PALMER[1,2], L. M. SAKSIDA[1,2], T. J. BUSSEY[1,2];
[1]Western Univ., London, ON, Canada; [2]Robarts Res. Inst., London, ON, Canada

Abstract: Background: Working memory (WM) is a cognitive process that enables individuals to temporarily store and manipulate information for decision-making and problem-solving. Various neuropsychiatric and neurodegenerative diseases are associated with WM deficits, which are often difficult to treat. To develop effective treatments, we need to understand the neurobiology underlying this cognitive process. Growing evidence supports the role of prefrontal cortex (PFC) parvalbumin (PV)-expressing neurons in attention and cognitive flexibility. However, findings related to the role of PV neurons in WM performance are inconclusive. **Objective:** To elucidate the role of PFC parvalbumin neurons in WM. **Methodology:** We used in vivo fibre photometry combined with a genetically encoded calcium sensor to record PV neuron calcium dynamics during a translational touchscreen trial unique nonmatch-to-location (TUNL) task that is used to assess spatial WM in both mice and humans. In this task, mice are required to remember the position of a stimulus presented on the screen over a delay period and select a different stimulus that is presented alongside the original stimulus. To record PV neuron activity in the PFC, PV-cre mice (n = 8; 3-6 months of age at the start of cognitive testing) were injected with AAV-syn-FLEX-jGCaMP7f-WPRE in the medial PFC (AP+1.8, ML+0.3, DV -2.2). A single optical fibre probe was implanted above the viral injection site. All mice were trained on the TUNL task prior to surgery and re-baselined on the task prior to fibre photometry recordings. Recordings were obtained during increasing delay durations across separate sessions (0s, 1s, and 2s) and during an interference probe in which the usual intertrial interval of 15s was removed. **Results:** Previously, our lab has demonstrated that optogenetic inhibition of prefrontal PV neurons during TUNL impairs task performance during higher cognitive load and increases susceptibility to interference. Consistent with these findings, greater PV neuron calcium activity were observed during the longer 2s delay and the interference probe, indicating differential recruitment of PV neurons depending on cognitive load. **Conclusions:** Our results show differential recruitment of PV neurons during WM task with increasing delays and when distracting information interferes with the target held in WM. These results indicate that a combination of translational touchscreen cognitive testing with cutting-edge imaging techniques, such as fibre photometry, can help us identify important neural targets for the treatment of cognitive deficits in patients with various neuropsychiatric and neurodegenerative conditions.

Disclosures: S. Hamidullah: None. **T.D. Dexter:** None. **D. Palmer:** None. **L.M. Saksida:** Other; Tim Bussey and Lisa Saksida have established a series of targeted cognitive tests for animals, administered via touchscreen within a custom environment known as the “Bussey-Saksida touchscreen chamber. **T.J. Bussey:** Other; Tim Bussey and Lisa Saksida have established a series of targeted cognitive tests for animals, administered via touchscreen within a custom environment known as the “Bussey-Saksida touchscreen chamber.

Poster

PSTR172. Learning and Memory: Physiology

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR172.10/RR7

Topic: H.08. Learning and Memory

Title: Parvalbumin-expressing neurons in perirhinal cortex mediate object discrimination and recognition.

Authors: *M. WOLTER[1,2], L. M. SAKSIDA[3], T. J. BUSSEY[3];
[1]Western Univ., Woodstock, ON, Canada; [2]Robarts Res. Inst., [3]Dept. of Physiol. & Pharmacol., Western Univ., London, ON, Canada

Abstract: It has been previously established in experiments in humans, non-human primates and rodents that the perirhinal cortex (PRh) is importantly involved in the visual discrimination and recognition of complex objects. These experiments were driven by predictions from computational models in which lateral inhibition in the cortex tunes cortical representations and provides a familiarity/novelty signal allowing discrimination and recognition of novel and familiar objects. In the real brain, this lateral inhibition is provided by cortical interneurons. This putative mechanism has never been tested, however, due to a previous lack of ability to manipulate cortical interneurons selectively. In the present experiment we used optogenetic techniques to silence neurons in perirhinal cortex during an object recognition task in which the perceptual similarity of the objects was systematically manipulated. First, we found that silencing pyramidal neurons in perirhinal cortex during the sample (encoding) phase of the task with a negligible delay impaired the task when objects were similar, but not dissimilar, thus providing a replication-with-variation of our previous findings using lesions, thus validating the paradigm. Second, we found that silencing parvalbumin-expressing neurons produced the identical pattern of results. Finally, we tested this effect in a disease model, the 22q11.2 microdeletion model of neuropsychiatric diseases such as schizophrenia, which has been shown to have dysfunctional cortical parvalbumin-expressing interneurons. Consistent with the optogenetic findings, these mice were impaired on the task when objects were similar, but not dissimilar. These data support the idea that perirhinal cortical inhibition via parvalbumin-expressing interneurons is a critical mechanism underlying object discrimination and recognition.

Disclosures: M. Wolter: None. **L.M. Saksida:** None. **T.J. Bussey:** None.

Poster

PSTR172. Learning and Memory: Physiology

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR172.11/RR8

Topic: H.08. Learning and Memory

Support: CIHR Vanier Canada Scholarship
Canada First Research Excellence Fund (CFREF)

Title: Translating rodent touchscreen tasks to measure cognition in older adults at risk for type 2 diabetes

Authors: *O. R. GHOSH-SWABY[1], A. SCHNEIDER[2], D. PALMER[2], T. J. BUSSEY[4], L. S. NAGAMATSU[3], L. M. SAKSIDA[2];
[1]Neurosci., [3]Sch. of Kinesiology, [2]Western Univ., London, ON, Canada; [4]Robarts Res. Inst., London, ON, Canada

Abstract: Diabetes & older age are well-known risk factors for dementia. Indeed, there is evidence that older adults not diagnosed, but at risk for diabetes show early signs of cognitive decline, further exacerbated by excessive body weight or high blood glucose. Such a finding would have implications for early treatment strategies; however, the evidence is still sparse. We examined the correlation of risk factors for diabetes with cognitive function in older adults at risk for diabetes using a battery of touchscreen tasks translated from their rodent versions, as well as traditional pen-to-paper cognitive tests. Sixty male & female older adults between 60-80 years old who were at risk for diabetes (BMI ≥ 25 kg/m2, HbA1c ≥ 6.0%, & CANRISK score ≥ 21) completed 3 novel touchscreen tasks - paired associative learning (PAL; learning and object-in-location memory), progressive ratio (PR; motivation), & trial unique, non-matching to location (TUNL; spatial pattern separation & working memory). In addition, they were tested on pen-to-paper cognitive tests - MoCA (total cognitive score), trail-making (task switching), Stroop (selective inhibition), & digit span (working memory). A correlation analysis was performed between BMI or HbA1c and cognitive performance. Performance on touchscreen tasks was analyzed using a repeated measures one-way ANOVA with trial blocks, separation level, or delays as the independent variable & accuracy or time as the dependent variable. This population exhibited lower than average MoCA scores (25.23 ±1.90) when compared to clinical guidelines. Only a negative correlation was observed between HbA1c & backward digit recall of the digit span test (r^2=0.323, p=0.0004). There was no correlation between breakpoint and BMI or HbA1c during PR (r^2=0.323, p=0.0004). Interestingly, this population performed at chance level (59.7 ± 5.3% accuracy) on the PAL, indicating they were unable to learn object-location paired associates. This poor performance contrasts with that of young, healthy adult participants who achieved ~70% accuracy in the final trial block of PAL testing (Nithianantharajah et al., 2015). When manipulating the spatial similarity in distance between stimuli during TUNL, older adults at risk for diabetes were 10% lower in accuracy when stimuli were close together compared to further apart (p<0001). Participants also responded more slowly to stimuli at choice during TUNL when delays were 10 sec compared to 2 & 5 sec (p=0.003). Older adults at risk for diabetes exhibit decreased performance on tasks with higher demands on spatial pattern separation and working memory compared to lower, & are unable to acquire object-location paired associates.

Disclosures: O.R. Ghosh-Swaby: None. **A. Schneider:** None. **D. Palmer:** None. **T.J. Bussey:** None. **L.S. Nagamatsu:** None. **L.M. Saksida:** None.

Poster

PSTR172. Learning and Memory: Physiology

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR172.12/RR9

Topic: H.08. Learning and Memory

Support: NSERC Canadian Graduate Scholarship - Doctoral
CIHR Canadian Graduate Scholarship - Masters
BrainsCAN
Ontario Graduate Scholarship

Title: An automated touchscreen-based stimulus-response learning task for mice is sensitive to in vivo dopamine dynamics and synucleinopathy

Authors: *O. PRINCZ-LEBEL[1], H. RAI[1], M. SKIRZEWSKI[2], S. PANJWANI[1], V. NOVIKOV[1], A. ATTARAN[1], R. SANDOVAL CONTRERAS[1], A. CHU[1], C. LEMIEUX[1], C. TAN[1], V. F. PRADO[3], M. A. PRADO[1], P. A. MACDONALD[1], L. M. SAKSIDA[2], T. J. BUSSEY[4];
[2]Western Univ., [3]Univ. of Western Ontario/Robarts Res. Inst., [4]Robarts Res. Inst., [1]Western Univ., London, ON, Canada

Abstract: Strategies for routine behaviours, or habits, offer a quick and efficient way to make decisions, but they limit behavioural flexibility. Many psychiatric and neurodegenerative disorders are characterized by abnormal decision-making and dysfunctional habit formation, including Synucleinopathies like Parkinson's disease (PD). The habitual control of behaviour is known to be influenced by striatal neurocircuitry, which facilitates synaptic plasticity and strengthens stimulus-response (S-R) associations. One crucial neuromodulator that regulates activity in the striatum is dopamine, and a loss of modulatory control of striatal dopamine, as seen in PD, can impact habitual behaviours. It remains unclear, however, whether striatal-projecting DA afferents are *necessary for* or *guide* the acquisition and performance of S-R learning.
Here we uniquely combined automated touchscreen cognitive assessments, fibre photometry, genetically-encoded dopamine biosensors and inhibitory chemogenetics to selectively record and manipulate *in vivo* dopamine dynamics while mice performed the Visuomotor Conditional Learning Task - an established cognitive task which measures S-R learning. We report differential patterns of dopamine neuromodulation across the striatum throughout the acquisition and performance of S-R learning, and distinct behavioural consequences of blocking nigrostriatal or mesolimbic dopamine afferents. Furthermore, using a humanized mouse model and intra-cranial injections of alpha-synuclein pre-formed fibrils, a key pathophysiological hallmark of PD, we highlight the translational potential of our S-R learning task and recommend its use for future preclinical or co-clinical trials.

Disclosures: **O. Princz-Lebel:** C. Other Research Support (receipt of drugs, supplies, equipment or other in-kind support); StressMarq Biosciences. **H. Rai:** None. **M. Skirzewski:** None. **S. Panjwani:** None. **V. Novikov:** None. **A. Attaran:** None. **R. Sandoval Contreras:** None. **A. Chu:** None. **C. Lemieux:** None. **C. Tan:** None. **V.F. Prado:** None. **M.A. Prado:** None. **P.A. MacDonald:** None. **L.M. Saksida:** None. **T.J. Bussey:** None.

Poster

PSTR172. Learning and Memory: Physiology

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR172.13/RR10

Topic: H.08. Learning and Memory

Support: BrainsCAN. Canada First Research Excellence Fund
CIHR, PJT 162431, PJT 159781
402524-2013 RGPIN; 03592-2021 RGPIN

Title: Awry acetylcholine-dopamine interaction within the nucleus accumbens of mice leads to impaired associative learning

Authors: *M. SKIRZEWSKI[1], O. PRINCZ-LEBEL[2], A. CROOKS[2], L. GERMAN-CASTELAN[2], G. KYUNGWOOK KIM[2], S. HENKE TARNOW[2], A. C. REICHELT[2], M. JING[4], S. MEMAR[2], F. SUN[5], Y. LI[6], L. M. SAKSIDA[3], V. F. PRADO[7], M. A. PRADO[8], T. J. BUSSEY[9];
[1]MouseTRAP, [3]Western Univ., [2]Western Univ., London, ON, Canada; [4]Chinese Inst. for Brain Research, Beijing, Beijing, China; [5]UCLA, UCLA, Los Angeles, CA; [6]Peking Univ., Peking Univ., Beijing, China; [7]Univ. of Western Ontario/Robarts Res. Inst., Univ. of Western Ontario/Robarts Res. Inst., London, ON, Canada; [8]Robarts Res. Institute/University of Western O, Robarts Res. Institute/University of Western O, London, ON, Canada; [9]Robarts Res. Inst., Robarts Res. Inst., London, ON, Canada

Abstract: Learning to associate environmental cues and rewards is fundamental for survival. Psychiatric disorders displaying abnormal associative learning endophenotypes have been correlated with altered local network circuitry mechanisms within the nucleus accumbens (NAc). Striatal cholinergic interneurons (CINs) are the main source of acetylcholine (ACh) in the striatum and play a robust neuromodulatory role onto dopaminergic axons, glutamatergic excitatory inputs, and synaptic plasticity on spiny projecting neurons (SPNs). Moreover, CINs are considered key players in reward-related learning behaviors because during salience, neurons respond with brief pauses, flanked by bursts of increased activity. Notwithstanding, it is still poorly described how altered ACh release from CINs affects NAc dopamine (DA) function and SPN activity in mice performing a reward-based associative learning task. We combined the use of automated touchscreens to assess a classic conditioning pavlovian task, transgenic mice harboring deficits in the expression of the vesicular ACh transporter (VAChT) in CINs (D2-cre,VAChT$^{f/f}$), and fiber photometry to record NAc ACh, DA, and calcium dynamics from SPNs. We show that mice with impaired ACh release from CINs leads to 1) deficits at discriminating cue stimuli predicting rewards, 2) impaired DA signal-to-noise ratio dynamics between CS+/CS- stimuli, 3) abnormal D1-SPN/D2-SPN calcium dynamics, and 4) these behavioral and NAc circuitry deficits are rescuable by bilateral expression of VAChT in NAc CINs via custom-made AAV-VAChT infection. Our findings suggest that ACh release from NAc CINs is fundamental to regulate local network circuitry underlying associative learning behaviors, yet deficits can be rescued during adulthood.

Disclosures: **M. Skirzewski:** None. **O. Princz-Lebel:** None. **A. Crooks:** None. **L. German-Castelan:** None. **G. Kyungwook Kim:** None. **S. Henke Tarnow:** None. **A.C. Reichelt:** None. **M. Jing:** None. **S. Memar:** None. **F. Sun:** None. **Y. Li:** None. **L.M. Saksida:** None. **V.F. Prado:** None. **M.A. Prado:** None. **T.J. Bussey:** None.

Poster

PSTR172. Learning and Memory: Physiology

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR172.14/RR11

Topic: H.08. Learning and Memory

Support: NSERC Grant R5476A06
NSERC CGS-M Award

Title: Development and pharmacological validation of a touchscreen operant task of cognitive judgement bias in mice

Authors: *A. Y. HERSEY, R. KIM, P. A. S. SHEPPARD, D. PALMER, T. J. BUSSEY, L. M. SAKSIDA;
Western Univ., London, ON, Canada

Abstract: Cognitive judgement bias (CJB) refers to the interpretation of ambiguous stimuli in a negative (pessimistic) or positive (optimistic) way. Negative CJB can be observed in depression, anxiety, and chronic illness, which can hinder the lives of individuals and caregivers. Animal models are critical for understanding and treating the cognitive symptoms of these neurological conditions; therefore, tasks assessing CJB in animals are essential for understanding the neural basis of CJB and for developing treatments. Many tasks used to assess CJB in animals have not been adequately validated or use aversive methodology that can confound treatments and compromise translational validity. This stands in contrast to human tests which are often computer-automated, in combination with touchscreens. Thus, our objective was to create a non-stressful touchscreen-based task for mice. We assessed CJB in C57BL/6J mice (8 male, 7 female) using an adaptation of a previously used touchscreen-based task (Lopez-Cruz et al., in prep), with a new set of stimuli and modified parameters. Stimuli were circular cues with diagonal lines rotated to varying degrees. Mice 3 months old at training onset were trained to respond to one rewarded cue (e.g., diagonal lines 45^{o} left) and withhold response from another unrewarded cue (e.g., diagonal lines 45^{o} right). During testing probes, we presented animals with 3 ambiguous, untrained cues (e.g., 22.5^{o} left, vertical (100% ambiguous) or 22.5^{o} right) dispersed among the trained cues, and calculated hit rate for each cue. Following baseline measurements (without intervention), we administered bupropion (5-10 mg/kg IP), or tetrabenazine (6 mg/kg IP) which are known to induce positive and negative CJB, respectively, with vehicles in an experimenter-blinded, Latin-square design. A priori paired t-tests revealed a significant decrease in hit rate to the 100% ambiguous cue (i.e. vertical lines), with tetrabenazine treatment, indicating a shift to negative CJB ($p < 0.01$). Conversely, 10 mg/kg bupropion resulted in a

significant increase in hit rate to the 100% ambiguous cue ($p < 0.01$), indicating positive CJB. These results provide a validated task to assess CJB in mice, which will allow us to ask future questions surrounding its underlying neurobiology and its relationship to, e.g., stress, other aspects of cognition, and treatments including lifestyle-based interventions.

Disclosures: **A.Y. Hersey:** None. **R. Kim:** None. **P.A.S. Sheppard:** None. **D. Palmer:** None. **T.J. Bussey:** Other; Campden Instruments. **L.M. Saksida:** Other; Campden Instruments.

Poster

PSTR172. Learning and Memory: Physiology

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR172.15/RR12

Topic: H.08. Learning and Memory

Support: BBRF NARSAD 29250
SNU startup

Title: Novelty selectively permits learning-associated plasticity in ventral tegmental-hippocampal-prefrontal circuitry

Authors: *A. PARK;
Physiol., Seoul Natl. Univ. Col. of Med., Seoul, Korea, Republic of

Abstract: The ability to modify established behavior in novel situations is essential, and patients with neuropsychiatric disorders often lack this flexibility. Understanding how novelty affects behavioral flexibility therefore has therapeutic potential. We designed a behavioral paradigm to test whether novelty exposure facilitates behavioral flexibility. Male mice ran free choice sessions in which they can freely choose an arm of a T-maze to get a reward for 3 days. As a result, mice developed an efficient strategy to simply choose one particular side approximately 90 % of the time (arm bias). The following day, mice were exposed to either a novel or familiar arena and trained on a delayed-non-match-to-sample flexible choice task. Mice had to learn to overcome their established bias and flexibly choose an arm opposite to the sample arm. We previously reported that novelty facilitates this flexible learning by weakening existing connectivity in the ventral hippocampal-medial prefrontal (vHPC-mPFC) circuit. This allows learning-induced connectivity re-strengthening during subsequent flexible choice training in a dopamine D1-receptor (D1R)-dependent mechanism (Park et al. Nature 2021). Here, to further investigate whether novelty induces circuit plasticity, the present study examined simultaneous recordings from the HPC, mPFC, and ventral tegmental area (VTA). Circuit connectivity was measured by local field potential (LFP) coherence. As mice exposed to novelty learned to overcome previously established spatial bias, the vHPC strengthens its coherence with the VTA and mPFC in theta frequency (4-8 Hz). Novelty or learning did not affect circuits involving the dorsal HPC. Without novelty, however, mice continued following established spatial bias and connectivity strength remained stable in the VTA-HPC-mPFC circuit. Pharmacologically

blocking D1Rs in the vHPC abolished the behavioral and physiological impacts of novelty. Thus, novelty promotes behavioral adaptation by permitting learning-associated plasticity in the vHPC-mPFC and VTA-vHPC circuit, a process mediated by D1Rs in the vHPC.

Disclosures: A. Park: None.

Poster

PSTR172. Learning and Memory: Physiology

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR172.16/RR13

Topic: D.06. Vision

Support: NIH Grant R01MH116500

Title: Delayed working paradigm in a freely moving environment shows deficits in the learning of FX mice

Authors: *S. NAREDDULA, R. MOFFITT, V. SALDARRIAGA, R. RUDNICKI, A. A. CHUBYKIN;
Biol. Sci., Purdue Univ., West Lafayette, IN

Abstract: Autism spectrum disorder (ASD) is a neurodevelopmental disorder that widely affects information processing in the brain resulting in deficits in learning and memory. One of the most prevalent forms of ASD is Fragile X Syndrome (FXS), which results from a mutation in the FMR1 protein. Previous studies have shown alterations in cell morphology, synaptic connections, and neural circuits pertaining to sensory perception in FXS model systems. Consistent with this, our lab has identified significant differences in the visual response of FX mice to a passive visual perceptual experience paradigm, specifically regarding evoked low-frequency theta (4-8 Hz) oscillations in the primary visual cortex (V1). These oscillations were attenuated in duration, amplitude, and frequency, suggesting these oscillations are a possible mechanism for visual working memory, and their impairment leads to a learning disability. However, currently, there is no widely accepted working memory behavior paradigm in mice. Here, we describe a new modified working memory paradigm based on a classical go/no-go visual discrimination task in mice. By utilizing ABETT touchscreen chambers and requiring the mice to wait for a period of time following a visual stimulus before responding in both wild-type and FX mice, we validated this new method and characterized the learning disability in FX mice. We found that across the multiple behavior paradigms, both WT and FX mice were able to show proper discrimination of the visual stimuli. However, we found that the FX mice consistently required more training days and reached lower overall training scores compared to the WT. Additionally, using DeepLabCut software we discovered that FX mice demonstrated distinct movement patterns consistent with impaired memory during freely moving behavior. Our findings highlight the efficacy of this novel method for studying working memory in mice. By employing this approach, we can gain deeper insights into the specific impairments associated

with FX mice, shedding light on the underlying mechanisms and potential therapeutic targets for addressing working memory deficits in this neurodevelopmental disorder.

Disclosures: S. Nareddula: None. **R. Moffitt:** None. **V. Saldarriaga:** None. **R. Rudnicki:** None. **A.A. Chubykin:** None.

Poster

PSTR173. Pharmacology of Learning and Memory

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR173.01/RR14

Topic: H.08. Learning and Memory

Support: NIH Grant NS072179
NIH Grant NS111389
NIH Grant NS126418

Title: Neuromodulator Orexin Fine Tunes the Physiological Substrates of Learning and Memory

Authors: *G. LATONA, J. A. KOSTANSEK, IV, S. MATTHEWS, K. A. SIMEONE, T. A. SIMEONE;
Dept. of Pharmacol. and Neurosci., Creighton Univ., Omaha, NE

Abstract: Orexins are excitatory neuropeptides produced in the lateral hypothalamus. There are two forms of orexin, orexin-A and orexin-B, that bind to two different subtypes of G-protein coupled receptors: orexin receptor-1 (OX_1R) and orexin receptor-2 (OX_2R). Orexins are implicated in regulating many different biological processes including sleep-wake cycles, feeding behaviors, among others. Behavioral studies of high orexin models show impaired performance on learning and memory tasks. Thus, orexin may be important in learning and memory, but the neural mechanism is not fully understood. This study aims to elucidate this neural mechanism. Multi-electrode array electrophysiology was used to determine the effects of orexin on SPW-ripple complexes and LTP in hippocampal slices *ex vivo* at baseline and following bath application of both the dual-orexin receptor antagonist N-[1,1'-Biphenyl]-2-yl-1-[2-[(1-methyl-1H-benzimidazol-2-yl)thio]acetyl-2-pyrrolidinedicarboxamide (TCS-1102; TCS) and the OX_1R antagonist N-(2-Methyl-6-benzoxazolyl)-N'-1,5-naphthyridin-4-yl urea (SB-334867; SB). TCS treatment significantly decreased SPW frequency while SB treatment significantly decreased SPW frequency, amplitude, and duration. In addition, TCS decreased ripple frequency and duration while SB only decreased the frequency of ripple events. Conversely, TCS treatment has limited impacts on LTP and SB's effects on LTP are still to be determined in future experiments. These results suggest that the neuromodulator orexin contributes to shaping the spontaneous SPW-ripple complexes, specifically by permitting more events of larger magnitude, which are associated with memory consolidation. In addition, these results suggest that while orexin may be involved in SPW-ripple complexes in the CA1, it may play a lesser role in LTP generation in the CA3-CA1 synapse of the hippocampus.

Disclosures: G. Latona: None. **J.A. Kostansek:** None. **S. Matthews:** None. **K.A. Simeone:** None. **T.A. Simeone:** None.

Poster

PSTR173. Pharmacology of Learning and Memory

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR173.02/RR15

Topic: H.08. Learning and Memory

Title: Effect of serotonin 1B/1A receptor activation on probabilistic reversal learning

Authors: *D. AMODEO, S. LOPEZ, M. GONZALES, M. JURADO, L. AMODEO; California State Univ. San Bernardino, San Bernardino, CA

Abstract: Serotonin 1B/1A receptors (5-HT1B/1A) have been known to impact learning and cognition. Previous studies have found that modulation of these receptors can enhance performance on reversal learning tasks. Reversal learning is often utilized to gage behavioral inflexibility, a symptom present in several neuropsychiatric disorders. Our previous study found that the 5-HT1B/1A receptor agonist RU24969, impaired probabilistic reversal performance and working memory in C57BL/6J mice. The current study aimed to examine the reproducibility of our 5-HT1B/1A receptor modulation findings in the spatial T-maze in mouse operant chambers. We also examined the potential sex differences that RU24969 has on probabilistic reversal learning. In addition, the current study utilized repeated drug administration compared to the single acute treatments used in our spatial tests. We predicted that the operant chamber results will parallel results from the spatial tests, inducing behavioral inflexibility in both males and females after RU24969 treatment. Mice were tested in operant discrimination task using an 80/20 probabilistic reinforcement procedure similar to those used in the spatial task. Mice were tested on a two-choice operant task, each mouse was tested on acquisition and then reversal learning across several days. Mice learned to obtain a sucrose pellet reinforcement from the magazine once a “correct” nose poke was made (reinforced on 80% of trials) compared with the “incorrect” nose poke (reinforced on 20% of trials). During each reversal day, mice received an intraperitoneal injection of either 0, 1.0, or 5.0 mg/kg of RU24969 10 minutes prior to each session. Both males and females displayed increased trials and days to reach learning criterion for reversal learning. The vehicle treated female C57BL/6J mice required more days and trials to reach criterion during reversal compared to vehicle treated male C57BL/6J mice. Male mice that received 1.0 mg/kg RU24969 displayed comparable reversal learning performance while 5.0 mg/kg impaired reversal learning performance.

Disclosures: D. Amodeo: None. **S. Lopez:** None. **M. Gonzales:** None. **M. Jurado:** None. **L. Amodeo:** None.

Poster

PSTR173. Pharmacology of Learning and Memory

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR173.03/RR16

Topic: H.08. Learning and Memory

Support: Wellcome Trust, UK
Epilepsy Research, UK

Title: Neurosteroid Pregnenolone Sulphate restores cognitive impairment and reduces ictal activity in a neuronal antibody-mediated seizure rat model

Authors: *M. UPADHYA[1], S. GANDHI[1], I. DIAS[1], S. R. IRANI[2], H. PRÜSS[3], S. BARMAN[4], N. GOEBELS[4], G. WOODHALL[1], S. WRIGHT[1];
[1]Aston Univ., Birmingham, United Kingdom; [2]Nuffield Dept. of Clin. Neurosciences, Univ. of Oxford, Oxford, United Kingdom; [3]4German Ctr. for Neurodegenerative Dis. (DZNE) Berlin and Charité Universitätsmedizin, Berlin, Germany; [4]Dept. of Neurology, Med. Faculty, Heinrich Heine Univ. Dusseldorf, Dusseldorf, Germany

Abstract: Background: NMDAR-Ab mediated encephalitis is a neuro-immunological disorder that presents with neuropsychiatric symptoms, seizures and movement disorder. NMDAR-Abs exert their pathogenic effect by NMDAR internalisation. The neurosteroid Pregnenolone sulfate (PregS) increases cell-surface expression of NMDARs. Our previous ***in vitro*** studies show that NMDAR-Ab-mediated ictal activity is reduced by PregS (Wright et al., 2021). Herein, we aimed to determine ***in vivo*** effects of PregS injections on ictal activity and behaviour in our NMDAR-Ab-mediated seizure rodent model. **Method:** PregS levels were determined from rat brain tissue (0.1 mg) spiked with internal standard (1ng of pregnenolone-17α,21,21,21-d4 sulfate) using LC-MS method. For EEG and behavioural studies, rats were divided into four groups: Control-Ab (n=8), Control-Ab+PregS (n=7), NMDAR-Ab+ vehicle (cyclodextrin) (n=7) and NMDAR-Ab+PregS (n=6) treated. The rats received continuous intracerebroventricular infusion of control-Ab or NMDAR-Ab for 7 days and in-vivo EEG was recorded until day 9. Rats in treatment groups were injected daily with CDX/PregS via subcutaneous routes. Novel Object Recognition (NOR), distance travelled and velocity were measured on days 1, 5 and 9 following surgery. Ictal activity was analysed using Neuroarchiver software (Opensource Instruments). **Results:** Endogenous PregS levels were observed in control and CDX brain samples and increased following PregS injection. On day 5 behavioural tests the NOR index was reduced in the NMDAR-Ab+vehicle treated rats as compared to the control-Ab group ($p<0.05$). This cognitive impairment was prevented in the NMDAR-Ab infused rats treated with PregS ($p<0.05$). Distance travelled and velocity was significantly decreased in NMDAR-Ab-vehicle as compared to Control-Ab, but not in NMDAR-Ab-PregS treated. Evolution of ictal events was reduced in the NMDAR-Ab-PregS treated rats as compared to NMDAR-Ab-vehicle treated rats. **Discussion and Conclusion:** Rodent brain PregS levels are increased following subcutaneous injections. *In vivo* treatment with PregS, a positive NMDAR modulator, restores cognitive dysfunction, prevents anxiety-like behaviour, and ictal activity in an NMDAR-Ab mediated rat seizure model. This proof-of-concept study highlights the potential use of receptor specific

treatments in antibody mediated epileptic encephalopathy.Reference: Wright S, et al. Commun. Biol. 2021;4:1106.

Disclosures: **M. Upadhya:** None. **S. Gandhi:** None. **I. Dias:** None. **S.R. Irani:** None. **H. Prüss:** None. **S. Barman:** None. **N. Goebels:** None. **G. Woodhall:** None. **S. Wright:** None.

Poster

PSTR173. Pharmacology of Learning and Memory

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR173.04/Web Only

Topic: H.08. Learning and Memory

Support: VIEP-BUAP 2023
CONACYT CF-2023-G-597

Title: Jwh-133 administration does not improve metabolic profile neither spatial memory in the obese zucker rat

Authors: *D. MANUEL SÁNCHEZ[1], E. MARTÍNEZ-PÉREZ[3], D. VALENCIA[3], A. PATRICIO-MARTÍNEZ[1], B. SILVA-GÓMEZ[4], I. LIMÓN[3], *D. MANUEL SÁNCHEZ[2]; [1]Lab. of Neuropharmacology, Fac. of Chem. Sci., Benemérita Univ. Autónoma De Puebla, Puebla, Mexico; [2]Benemérita Univ. Autónoma De Puebla, Amozoc, Mexico; [3]Lab. of Neuropharmacology, Fac. of Chem. Sci., [4]Lab. of Exptl. Neurophysiology, Fac. of Biol. Sci., Benemérita Univ. Autónoma de Puebla, Puebla, Mexico

Abstract: Obesity is defined as the excessive fat accumulation and is linked whit the development of chronic diseases such as type II diabetes mellitus. Several studies have shown that obesity affects glutamatergic communication in the hippocampus and impairs spatial memory. For this reason, new therapeutic alternatives are being explored to control obesity as well as improving the cognitive impairment it causes. In this context, the CB2 receptor has acquired significant relevance since it has been shown to modulate energy balance and synaptic plasticity. The aim of the present study was to evaluate the effect of chronic activation of the CB2 receptor on metabolic homeostasis and spatial memory in the obese Zucker rat. We used 10 LZDF (Lean Zucker Diabetic Fatty) rats as the control group and 8 OZDF (Obese Zucker Diabetic Fatty) rats as the experimental group. At 12 weeks of age we assessed glucose homeostasis through glucose tolerance test and spatial learning in the Morris water maze, where we obtained escape latency and navigation maps. After the last learning assay, the initial groups were subdivided into 2 groups and for 10 consecutive days a dose of 0.2 mg/kg of JWH-133 (CB2 receptor agonist) or vehicle was intraperitoneally administered. In the spatial memory we assessed the following variables in the platform zone: latency to first entry, number of entries and time spent. In addition, we monitored body weight and determined serum levels of glucose, cholesterol and triglycerides and the expresión of CB2 receptor in CA1 hippocampus area. Our results showed that OZDF rats show an obese phenotype that is concomitant with metabolic

dysregulation in relation whit increases in glucose, cholesterol and up to 3-fold increase in triglyceride levels compared to LZDF rats. Furthermore, this animals showed learning and memory impairment, while JWH-133 administration did not improve their lipid and glycaemic profile, nor their performance in the spatial memory test and CB2 receptor expression.In conclusion, our work shows that obesity promotes learning delay and spatial memory deficits possibly through neuroinflammation, whereas administration of JHW-133 through 10 days is not sufficient to ameliorate the metabolic profile and spatial memory deficits in the OZDF rats.

Disclosures: **D. Manuel Sánchez:** None. **E. Martínez-Pérez:** None. **D. Valencia:** None. **A. Patricio-Martínez:** None. **B. Silva-Gómez:** None. **I. Limón:** None. **D. Manuel Sánchez:** None.

Poster

PSTR173. Pharmacology of Learning and Memory

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR173.05/RR17

Topic: H.08. Learning and Memory

Support: VIEP-BUAP 2023 (BUAP-CA-288)
LD PhD CONACYT Grant No. 850282

Title: Corticosterone influences differentially the memory retrieval of innate anxious and resilient rats

Authors: *L. DÍAZ[1], A. UGARTE[2], C. CORTES[3], J. R. EGUIBAR[4];
[1]Benemérita Univ. Autónoma De Puebla, Heroica Puebla de Zaragoza, Mexico; [2]Inst. de Fisiología, [4]Intl. Office, [3]Benemérita Univ. Autónoma de Puebla, Puebla, Mexico

Abstract: Usually during learning the subject have a stressful condition that increase the release of stress hormones and then enhanced memory consolidation. In contrast, the administration of exogenous glucocorticoids like corticosterone, shortly before memory tests, impairs the retrieval phase, when done in subjects with average stress responses. However, less is known about the effects of these hormones in the memory of innate anxious or resilient subjects. We selectively inbreed two sublines from Sprague-Dawley (SD) rats, the first with a high-spontaneous yawning (HY) with a mean of 20 yawns/h, and low-yawning (LY) frequency with just 2 yawns/h. When tested in the open-field arena LY rats ambulated less and had more fecal bolus with respect to HY males, showing a higher stress response the former. Furthermore, LY subline, evaluated in the Barnes maze, showed deficits with respect to HY in long-term memory. So, the aim of this study was to evaluate the effects of exogenous corticosterone administration on memory retrieval in both sublines. We used LY and HY male rats (6 rats/group), they were maintained under standard conditions. All experiments were done between 1000 to 1300 h using a Barnes maze. The rats trained two consecutive days with four trials, in every trial subjects (Ss) learned a fixed position of the escape box with extra-maze cues available. Seven days later and 30 minutes before long-term memory (LTM) retrieval, Ss received 0.3, 1.0 or 3.0 mg/Kg of corticosterone

diluted in a physiological saline solution with 5% ethanol (Sigma-Aldrich, USA), injected subcutaneously in the dorsal neck region. The escape latency and number of errors were measured. Our results showed that 1 mg/Kg of corticosterone increased the number of errors in HY, and decreased it in the LY subline, with respect to control group ($P<0.05$). The escape latencies increased in SD and LY groups with 0.3 mg/Kg of corticosterone during long-term memory evaluation, but this dose did not have effect in HY rats ($P<0.05$). We concluded that corticosterone had opposite effects in LTM retrieval on HY and LY sublines, probably due to differences in glucocorticoid receptor expression in limbic structures like the hippocampus and basolateral amygdala.

Disclosures: L. Díaz: None. **A. Ugarte:** None. **C. Cortes:** None. **J.R. Eguibar:** None.

Poster

PSTR173. Pharmacology of Learning and Memory

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR173.06/RR18

Topic: H.08. Learning and Memory

Support: NIH Grant NS128039

Title: Enhancing or suppressing GIRK channel activity in the dorsal hippocampus triggers cognitive impairment in mice

Authors: *M. FREDERICK[1], H. LUO[2], E. MARRON[2], J. R. OSTERLUND OLTMANNS[2], C. WRIGHT[2], K. D. WICKMAN[3];
[1]Pharmacol., Univ. of Minnesota, Twin Cities, Minneapolis, MN; [2]Univ. of Minnesota, Minneapolis, MN; [3]Dept Pharmacol, Univ. Minnesota, MINNEAPOLIS, MN

Abstract: A balance of excitatory (E) and inhibitory (I) neurotransmission is critical for neuronal function and synaptic plasticity. E/I imbalance in the dorsal hippocampus (dHPC), and in CA1 pyramidal neurons, has been implicated in the cognitive dysfunction associated with multiple neurological disorders, including Alzheimer's Disease and Down Syndrome. G protein-gated inwardly rectifying K+ (GIRK/Kir3) channels mediate the G protein-dependent postsynaptic inhibitory effects of many neurotransmitters, including GABA, serotonin, and adenosine, and exert a critical influence on the excitability of neurons in the hippocampus. While cognitive deficits have been reported in both gain- and loss-of-function models involving GIRK channels in mice, the anatomic and cellular basis of GIRK channel contributions to cognitive behaviors is unclear. Here, we examined the behavioral impact of strengthening or weakening GIRK channel activity in CA1 pyramidal neurons in the dHPC. We used TrpC4Cre transgenic mice to gain selective genetic access to CA1 pyramidal neurons and well-characterized Cre-dependent AAV vectors to drive selective enhancement (AAV-GIRK2) or suppression (AAV-GIRK3) of GIRK channel activity in CA1 pyramidal neurons in the dHPC. In one cohort of mice, we validated the impact of our manipulations using slice electrophysiological methods. A

separate cohort of mice was run through a behavioral battery consisting of light/dark exploration, Y-maze, open field activity, novel object recognition (NOR), and contextual fear conditioning (CFC) tests to assess the behavioral impact of our manipulations. We observed no main effect of sex or interaction between sex and viral treatment for any test; as such, data from male and female subjects were pooled. While no viral treatment effects were observed in light/dark exploration, open field, or Y-Maze tests, either enhancement or suppression of GIRK channel activity in CA1 pyramidal neurons in the dHPC provoked deficits in CFC ($F(2,10)=10.01$, $P=0.0041$; one-way ANOVA) and NOR ($F(2,18)=3.50$, $P=0.0519$; one-way ANOVA) tests. Thus, dysregulation of GIRK channel activity in CA1 pyramidal neurons in the dHPC leads to cognitive impairment in mice. Our work adds to a growing body of evidence that targeting GIRK channels may afford therapeutic benefits for neurological disorders characterized by cognitive deficits.

Disclosures: **M. Frederick:** None. **H. Luo:** None. **E. Marron:** None. **J.R. Osterlund Oltmanns:** None. **C. Wright:** None. **K.D. Wickman:** None.

Poster

PSTR173. Pharmacology of Learning and Memory

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR173.07/RR19

Topic: H.08. Learning and Memory

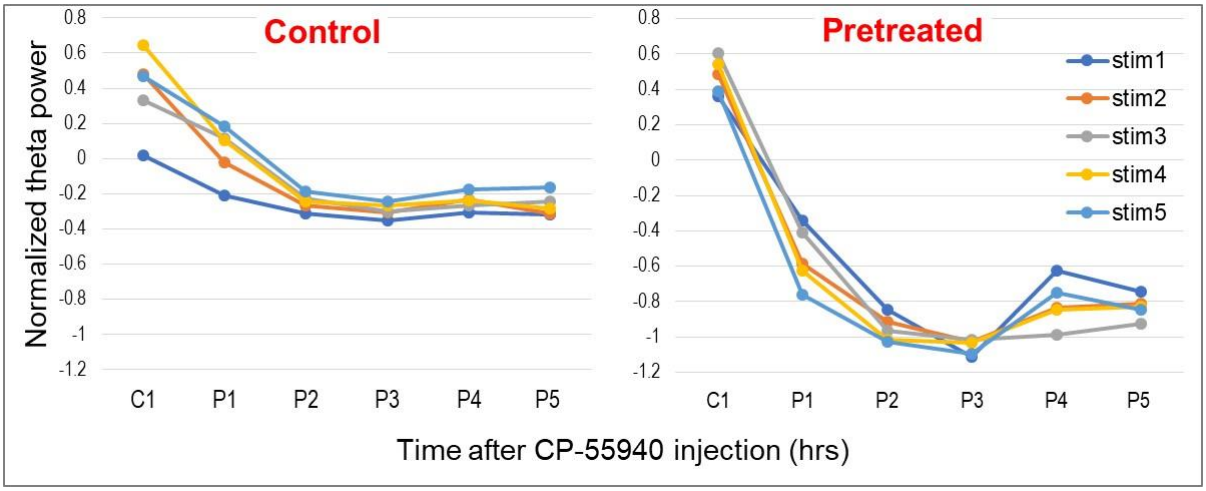
Support: MH127483

Title: The effect of Cannabinoid-1 receptor activation in adolescence on oscillatory networks in the hippocampus

Authors: J. REHN[1], *B. KOCSIS[2];
[1]Psychiatry-Beth Israel Med. Ctr., [2]Harvard Med. Sch., Boston, MA

Abstract: Activation of Cannabinoid receptors (CB1-R) was shown to interfere with neuronal network oscillations and to impair sensory gating function, supporting the connection between cannabis abuse and increased susceptibility of developing schizophrenia spectrum disorders. Historically, concern around adolescent marijuana use focused on the development of psychosis, however recent findings indicate that cognitive domains may also be at risk. Neuronal oscillations are essential in multiple cognitive functions and their impairment was documented in neurological and psychiatric diseases. The selective CB1-R agonist, CP-55940 significantly reduced theta and gamma power in the hippocampus (HPC) in freely behaving adult rats (Hajos et al. 2008). Disruption of slow oscillations in the theta range were also shown in a model using rats anesthetized with chloral hydrate and was reversed by the CB1-R antagonist AM-251. The potential involvement of CB1-R activation during adolescence however remains unexplored. This study pursued two goals: (1) to verify CB1-R agonist effect on HPC theta rhythm in the widely used model of urethane-anesthetized rats and (2) to investigate the long-term effect of

chronic treatment of rats by CP-55940 in early phases of development on their reaction to this compound once they reached adulthood. Rats were pretreated with chronic administration of CP-55940 (n=11, 6 males, 5 females) or vehicle (n=8, 4 males, 4 females) during adolescence (daily i/p injections between postnatal days PND32-36 or PND42-46, n=10 and 9). They were then tested in adulthood (PND70-88, n=17 or PND 111-115, n=2) under urethane anesthesia. HPC theta rhythm was elicited by brainstem stimulation at 5 levels of current intensity (see colors in Fig) one hour before (C1) and up to 5 hours after injection (P1-P5). We found a lasting significant decrease in theta power after CP-55940 in the urethane model (Fig., left) which was aggravated further in rats pretreated with the CB1-R agonist (Fig., right, theta power normalized to the lowest (=0) and highest (=1) values in control (C1) in each experiment).



Disclosures: **J. Rehn:** None. **B. Kocsis:** None.

Poster

PSTR173. Pharmacology of Learning and Memory

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR173.08/RR20

Topic: H.08. Learning and Memory

Support: P031S160068
5P20GM103642
SCORE-SC21SC2NS119144
R25GM110513

Title: An alpha 7 nicotinic acetylcholine receptor modulator, 4R-cembra-trienedienol, improves anxiety and spatial learning in Gulf War Illness male mice.

Authors: *J. MARRERO;
Neurosci., Univ. Central Del Caribe, San Juan, PR

Abstract: Gulf War Illness (GWI) is a chronic multiorgan condition primarily affecting the brain, characterized by persistent neuroinflammation, anxiety, and cognitive deficits. GWI causative factors are the simultaneous exposure to pyridostigmine bromide (PB, administered for prophylactic purposes), permethrin (PER), and N, N-diethyl-meta-toluamide (DEET) for insect control along with traces of Sarin. Despite the advancementsin this area, effective treatment against GWI has not been established. Here, we report the therapeutic efficacy of a novel neuroprotective compound and α7nAChR modulator, the 4R, against GWI. 4R is a non-toxic compound that crosses the blood brain barrier (BBB) and reaches higher concentrations in the brain than plasma. To recreate GW conditions, we administrated PB and PER with DEET, traces of DFP (a Sarin surrogate), and moderate stress for 12 consecutive days in C57BL/J6 mice. After 30 days of rest, which allowed the disease to express, we implemented an intraperitoneal (i.p.) 4R treatment: five times a week for four consecutive weeks (6mg/kg body weight). Our findings revealed that 4R improved spatial learning ability in GWI animals employing a two-day Barnes maze training protocol (training session: T1-T3 on day 1 and T4-T5 on day 2). The CON-VEH and GWI-4R group learned the location of the escape box by T3 with latency 79.38 ± 9.515 vs 77.15 ± 12.81 sec. Conversely, both GWI-VEH and CON-4R show insignificant learning improvement. During T1-5, the GWI-VEH group retained a base level of latency to escape box from T1 108.41 ± 7.863 sec to T5 97.40 ±9.333 sec. However, 48 hours later, all groups reached the escape hole with similar latency. Additionally, 4R improved anxiety-like behavior in GWI mice. The CON-VEH and CON-4R groups remained 50% longer in the Elevated Plus Maze's open arms compared to the GWI-VEH. Alternatively, the GWI-4R group spent less time in the closed arms and behaved similarly to the CON groups. Moreover, latency to open arms was decreased considerably in GWI after the 4R administration (GWI-VEH 91.83 ± 21.16 sec vs GWI-4R 14.95 ± 1.958 sec), suggesting that early onset of anxiety in GWI can be corrected via α7nAChR modulation. These results were not related to alterations in motor function measured by velocity and distance traveled. In conclusion, our study tested a promising candidate drug for treating GWI veterans and future victims of similar civilian or military adverse events.

Disclosures: **J. Marrero:** None.

Poster

PSTR173. Pharmacology of Learning and Memory

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR173.09/SS1

Topic: H.08. Learning and Memory

Title: Participation of M_2 muscarinic Acetylcholine receptors in the regulation of memory in rats during and after chronic REM sleep restriction @font-face {font-family:"Cambria Math"; panose-1:2 4 5 3 5 4 6 3 2 4; mso-font-charset:0; mso-generic-font-family:roman; mso-font-pitch:variable; mso-font-signature:-536870145 1107305727 0 0 415 0;}@font-face {font-family:Calibri; panose-1:2 15 5 2 2 2 4 3 2 4; mso-font-charset:0; mso-generic-font-family:swiss; mso-font-pitch:variable; mso-font-signature:-469750017 -1073732485 9 0 511 0;}p.MsoNormal, li.MsoNormal, div.MsoNormal {mso-style-unhide:no; mso-style-qformat:yes; mso-style-

parent:""; margin:0cm; margin-bottom:.0001pt; mso-pagination:widow-orphan; font-size:12.0pt; font-family:"Calibri",sans-serif; mso-ascii-font-family:Calibri; mso-ascii-theme-font:minor-latin; mso-fareast-font-family:Calibri; mso-fareast-theme-font:minor-latin; mso-hansi-font-family:Calibri; mso-hansi-theme-font:minor-latin; mso-bidi-font-family:"Times New Roman"; mso-bidi-theme-font:minor-bidi; mso-fareast-language:EN-US;}.MsoChpDefault {mso-style-type:export-only; mso-default-props:yes; font-family:"Calibri",sans-serif; mso-

Authors: S. NAVARRO-MONDRAGON, ***A. JIMENEZ-ANGUIANO**;
Biologia de la Reproduccion, Univ. Autonoma Metropolitana-Iztapalapa, Mexico City, Mexico

Abstract: It has been shown that sleep is involved in the consolidation and retrieval of declarative memory (DM). Acetylcholine is involved in the modulation of sleep and DM. However, it is still unknown the selective participation of muscarinic Acetylcholine receptors (mAChR) in both processes. The objective was to evaluate the effect of M_2 mAChR in the chronic REM sleep restriction (CREMSR) on DM, as well as during the recovery period of CREMSR through the novel object recognition test (NOR test). Eighteen male Wistar rats were used in the following groups (n=6): 1. Control, 2. CREMSR x 21 days, 3. CREMSR x 21 days + antagonist M_2 mAChR-Methoctramine (METHO 1 mg/kg i.p). CREMSR were performed using the multi-platform technique. At the beginning, at 11 and 21 days of the CREMSR and after 21 days during the recovery period the animals were evaluated in the NOR test. The results obtained showed that sleep deficit and METHO produced a decrease in NOR test. From the results obtained, we suggest that differential sleep loss and the blocking of M_2 mAChR have an impact in the correct learning and produces degradation in DM. @font-face {font-family:"Cambria Math"; panose-1:2 4 5 3 5 4 6 3 2 4; mso-font-charset:0; mso-generic-font-family:roman; mso-font-pitch:variable; mso-font-signature:-536870145 1107305727 0 0 415 0;}@font-face {font-family:Calibri; panose-1:2 15 5 2 2 2 4 3 2 4; mso-font-charset:0; mso-generic-font-family:swiss; mso-font-pitch:variable; mso-font-signature:-469750017 -1073732485 9 0 511 0;}p.MsoNormal, li.MsoNormal, div.MsoNormal {mso-style-unhide:no; mso-style-qformat:yes; mso-style-parent:""; margin:0cm; margin-bottom:.0001pt; mso-pagination:widow-orphan; font-size:12.0pt; font-family:"Calibri",sans-serif; mso-ascii-font-family:Calibri; mso-ascii-theme-font:minor-latin; mso-fareast-font-family:Calibri; mso-fareast-theme-font:minor-latin; mso-hansi-font-family:Calibri; mso-hansi-theme-font:minor-latin; mso-bidi-font-family:"Times New Roman"; mso-bidi-theme-font:minor-bidi; mso-fareast-language:EN-US;}.MsoChpDefault {mso-style-type:export-only; mso-default-props:yes; font-family:"Calibri",sans-serif; mso-ascii-font-family:Calibri; mso-ascii-theme-font:minor-latin; mso-fareast-font-family:Calibri; mso-fareast-theme-font:minor-latin; mso-hansi-font-family:Calibri; mso-hansi-theme-font:minor-latin; mso-bidi-font-family:"Times New Roman"; mso-bidi-theme-font:minor-bidi; mso-fareast-language:EN-US;}div.WordSection1 {page:WordSection1;}

Disclosures: S. Navarro-Mondragon: None. **A. Jimenez-Anguiano:** None.

Poster

PSTR173. Pharmacology of Learning and Memory

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR173.10/SS2

Topic: H.08. Learning and Memory

Support: RF1-AG060754

Title: Exploration of Deep Brain Stimulation effects on Neurotrophic Markers in Cerebrospinal Fluid (CSF) in aged Rhesus Macaque monkeys using an Ommaya

Authors: *D. BLAKE[1], **K. PENNINGTON**[2], F. VALE DIAZ[3], C. CRYAN[4];
[1]Med. Col. of Georgia at Augusta Univ., Augusta, GA; [2]Augusta Univ. Dept of Neurosci. & Regenerative Med., Augusta, GA; [3]Augusta Univ. Dept. of Neurosurg., Augusta, GA; [4]Augusta Univ. Neurosci. Grad. Program, Augusta, GA

Abstract: Neurotrophic biomarkers such as tissue-type plasminogen activator (tPA), plasminogen activator inhibitor-1 (PAI-1), and brain-derived neurotrophic factor (BDNF) have been identified as key players in the mechanisms of a multitude of disorders, including depression, dementia, post-traumatic stress disorder (PTSD), and many others. These are challenging to study in animal models, as they exist in cerebrospinal fluid (CSF) in pg/mL levels. Many common laboratory animals have too little CSF to create a sample for common assays like an enzyme-linked immunosorbent assay (ELISA) test. We developed a method that allows for increased volume of CSF collection by using the nonhuman primate model and collecting CSF through an Ommaya, or ventricular drain and reservoir. The Ommaya is placed into the base of one lateral ventricle in the nonhuman primates, and may safely sample 0.25 cc (250 μL) of CSF each 30 minutes. We used this sampling to assess whether deep brain stimulation of the nucleus basalis of Meynert leads to increases in CSF levels of tPA. Stimulation was intermittent at 60 pulses per second for 20 seconds on, 40 seconds off, for one hour. Pulses were monopolar, biphasic, cathodal first, 100 μS per phase at an amplitude of 0.5 mA, and were delivered for 20 seconds in each minute. In four aged macaque monkeys, we found tPA levels of 35-90 pg/mL at baseline, which increased 2-3 fold one hour after stimulation began. Future plans include assessing other potential neurotrophic biomarkers that also exist at similar levels in CSF.

Disclosures: D. Blake: None. **K. Pennington:** None. **F. Vale Diaz:** None. **C. Cryan:** None.

Poster

PSTR173. Pharmacology of Learning and Memory

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR173.11/SS3

Topic: H.08. Learning and Memory

Support: PhD scholarship for the first two authors from the Univesity of Western Australia (RTP Training)

Title: The Effects of Caffeine and D-amphetamine on Spatial Span Task in Healthy Participants

Authors: *F. M. KASSIM[1,2], M. LIM[4], S. SLAWIK[6], K. GAUS[6], B. PETERS[4], J. LEE[5], E. HEPPLE[5], J. RODGER[3], M. ALBRECHT[3], M. MARTIN-IVERSON[2];
[1]Psychiatry, St. Paul's Hosp. Millennium Med. Col., Addis Ababa, Ethiopia; [2]Pharmacol., [3]The Univ. of Western Australia, Perth, Australia; [4]Univ. of Western Australia, Nedlands, Australia; [5]Univ. of Western Australia, Perth, Australia; [6]Univ. of Bremen, Bremen, Germany

Abstract: Studies that examined the effect of amphetamine or caffeine on spatial working memory (SWM) and verbal working memory (VWM) have used various tasks. However, there are no studies that have used spatial span tasks (SSTs) to assess the SWM effect of amphetamine and caffeine, although some studies have used digit span tasks (DST) to assess VWM. Previous reports also showed that increasing dopamine increases psychosis-like experiences (PLE, or schizotypy) scores which are in turn negatively associated with WM performance in people with high schizotypy and people with schizophrenia. Therefore, the present study aimed to examine the influence of d-amphetamine (0.45 mg/kg, PO), a dopamine-releasing stimulant, on SST, DST, and on PLE in healthy volunteers. In a separate study, we examined the effect of caffeine, a nonspecific adenosine receptor antagonist with stimulant properties, on similar tasks.
Methods: Healthy participants (N=40) took part in two randomized, double-blind, counter-balanced placebo-controlled cross-over pilot studies: The first group (N= 20) with d-amphetamine (0.45 mg/kg, PO) and the second group (N=20) with caffeine (200 mg, PO). Spatial span and digit span were examined under four delay conditions (0, 2, 4, 8 s). PLE was assessed using several scales measuring various aspects of psychosis and schizotypy. **Results:** We failed to find an effect of d-amphetamine or caffeine on SWM or VWM, relative to placebo. However, d-amphetamine increased a composite score of psychosis-like experiences ($p = 0.0005$), specifically: Scores on the Brief Psychiatric Rating Scale, Perceptual Aberrations Scale, and Magical Ideation Scale were increased following d-amphetamine. The degree of change in PLE following d-amphetamine negatively and significantly correlated with changes in SWM, mainly at the longest delay condition of 8 s ($r = -0.58$, $p = 0.006$). **Conclusion:** The present results showed that moderate-high dose of d-amphetamine and moderate dose of caffeine do not directly affect performances on DST or SST. However, the results indicate that d-amphetamine indirectly influences SWM, through its effect on PLE.

Disclosures: F.M. Kassim: None. **M. Lim:** None. **S. Slawik:** None. **K. Gaus:** None. **B. Peters:** None. **J. Lee:** None. **E. Hepple:** None. **J. Rodger:** None. **M. Albrecht:** None. **M. Martin-Iverson:** None.

Poster

PSTR173. Pharmacology of Learning and Memory

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR173.12/SS4

Topic: H.08. Learning and Memory

Support: NIH Grant AG07823
NIH Grant AG056622

NIH Grant AG082388
A2017457S

Title: Investigation of the signaling mechanisms underlying the neuronal effects of metformin

Authors: *N. I. NICOL[1], H. JESTER[2], X. ZHOU[1], T. MA[3];
[1]Intrnl. Medicine-Geriatrics, Wake Forest Univ. Sch. of Med., Winston-Salem, NC; [2]Intrnl. Medicine-Geriatrics, Wake Forest Univ. Sch. of Med., Germanton, NC; [3]Intrnl. Medicine-Geriatrics, Wake Forest Sch. of Med., Winston Salem, NC

Abstract: Metformin is one of the most commonly prescribed drugs for treatment of diabetes. Recent studies in both clinical and basic science settings suggest that metformin treatment could improve cognitive impairments associated with aging and Alzheimer's disease (AD), although conflicting findings have also been reported. The molecular mechanisms underlying the neuronal effects of metformin remain unclear. Previous studies, mainly from non-neuronal cell culture systems, show that metformin functions as a potent activator of the AMP-activated protein kinase (AMPK) through the inhibition of the mitochondrial respiratory chain. This study aims to elucidate the signaling mechanisms of action of metformin in the brain. Acute hippocampal slices from ten-week-old, male C57BL6 mice were incubated in vehicle, 5 µM, or 100 µM of metformin for 1 hour. Western blot analysis was then performed to look at proteins related to the AMPK signaling pathway and the mitochondria. The data indicate a significant decrease in Sirtuin 1 (Sirt1) protein levels at both concentrations ($p = 0.01$ for 5 µM, $p = 0.04$ for 100 µM) and a trending increase in catalase protein levels at 5 µM of metformin ($p = 0.08$). Sirt1 is an NAD-dependent deacetylase involved in cellular regulation and catalase is an enzyme that breaks down hydrogen peroxide into water and oxygen. Such effects on Sirt1 and catalase regulation are in line with studies in liver tissue. Surprisingly, we did not observe any effect on AMPK activities (measured by phosphorylation of AMPKα at the T172 site) with treatment of metformin. Next, we will administer metformin to the mice via drinking water in either an acute or chronic dosing paradigm. The liver and brain tissues from these mice will be analyzed using to compare the potential mechanisms of action of metformin.

Disclosures: **N.I. Nicol:** None. **H. Jester:** None. **X. Zhou:** None. **T. Ma:** None.

Poster

PSTR174. Learning and Memory: Timing and Temporal Processing

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR174.01/SS5

Topic: H.08. Learning and Memory

Support: NIH Grant R01MH122391
NIH Grant U19NS107616
NSF grant 1707316

Title: Self-controlled acoustic-cue guided navigation: spatial and internally driven action signals in the hippocampus

Authors: *I. ZUTSHI[1], A. APOSTOLELLI[1], W. YANG[1], T. DOHI[1], E. BALZANI[2], C. SAVIN[2], G. BUZSAKI[3];
[2]New York Univ., [1]New York Univ., New York, NY; [3]New York Univ., New York University, Langone Med. Ctr., New York, NY

Abstract: While most often studied for their reliable spatial tuning, neurons within the hippocampus respond to other non-spatial features, especially when these cues are required to solve an ongoing task. However, it is unclear whether spatial and task-relevant cognitive tuning co-exist, compete, or cooperate within the hippocampus.
We implemented an agent-controlled '*acoustic cue-guided navigation task'* where the need for representing continuous change in space or other task variables (acoustic cue) was purposefully juxtaposed. Mice ran on a linear track with 7 equally spaced ports. During non-auditory trials, mice received a water reward at either end of the track. During auditory trials, tone frequencies ascended in closed loop to the spatial position of mice. The sweep speed was varied such that across trials the same frequency coincided with different spatial locations. The mice must identify when the target frequency (22 kHz) was reached and lick at the closest water port. Recordings from CA1 neurons revealed a similar fraction of place cells in non-auditory and auditory trials, despite the spatial variable being irrelevant for the auditory task. Thus, spatial tuning persisted. In addition, ~15% of all recorded cells were tuned to the auditory frequency. Such auditory tuning resulted in distinct spatial fields from trial-to-trial, depending on the mouse's choice. Using a combination of non-auditory trials and behavioral controls, we found that most of these non-spatial 'auditory' responses were driven by the animal's internally generated signals for upcoming actions, rather than reflecting purely auditory or reward tuning. To effectively quantify how cells were tuning to spatial or task-relevant variables, we fit the firing of single neurons to multiple external variables by using a Poisson Generalized Additive Model (Balzani et al., 2020) to classify cells as spatial or task-relevant.
We next used dimensionality reduction methods to dissect how spatial and task-relevant cells cooperate at the population level. Similar to the single cell level analysis, we observed that the neural manifold represented spatial location but also segregated to predict the mouse's exact choice during tone trials. Excluding task-selective neurons identified by the PGAM did not disrupt the spatial representations, but decreased choice location-based segregations on the manifold.
Together, these results indicate that hippocampal firing combines spatial and internally generated cognitive variables relevant for solving the task. These tuning properties cooperate to support context-specific representations that reflect ongoing choices and actions in the hippocampus.

Disclosures: I. Zutshi: None. **A. Apostolelli:** None. **W. Yang:** None. **T. Dohi:** None. **E. Balzani:** None. **C. Savin:** None. **G. Buzsaki:** None.

Poster

PSTR174. Learning and Memory: Timing and Temporal Processing

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR174.02/SS6

Topic: H.08. Learning and Memory

Title: Mechanisms of hippocampo-cortical interaction during NREM sleep

Authors: *R. SWANSON[1], J. BASU[2], G. BUZSAKI[3];
[1]New York Univ., New York, NY; [2]Dept. of Neurosci. and Physiol., Neurosci. Institute, New York Univ. Sch., New York, NY; [3]Neurosci., New York University, Langone Med. Ctr., New York, NY

Abstract: During NREM sleep, the brain is in a self-organized excitable regime in which alternations between spiking and near cessation of spiking propagate along the forebrain, termed slow oscillations (SOs) or UP and DOWN states in the neocortex, and sharpwave-ripples (SPW-Rs) in the hippocampus (Levenstein 2019). Both gain and loss of function studies have demonstrated the importance of tight temporal coordination between SOs and SPW-Rs systems consolidation. However, where, when, and how this tight coupling is spontaneously achieved across regions is unknown, despite being essential for understanding whole-brain mechanisms of systems consolidation. Towards this goal, we developed a chronic preparation in mice that combines widefield imaging of dorsal neocortex and ipsilateral extracellular silicon probe recordings of hippocampus (HPC) and retrosplenial cortex (RSC), allowing us to monitor multi-scale interaction between regions during sleep and further develop an existing theory of NREM sleep.
We find that interaction between HPC and RSC is well matched by a proposed model whereby both RSC and HPC are in reciprocally perturbable excitable regimes, and the degree to which they can perturb one another depends on both the strength of input received and state of the receiving region. More specifically, 1. SPW-Rs can cause DOWN states in RSC that may propagate to lower order visual areas conditional on the magnitude of the SPW-R and the state of cortex, 2. transitions to UP states are sufficiently synchronous to drive SPW-Rs in hippocampus, and 3. SPW-Rs occurring during DOWN states are uniquely poised to influence spike latencies at the DOWN to UP transition, suggesting a putative mechanism for consolidation.

Disclosures: R. Swanson: None. **J. Basu:** None. **G. Buzsaki:** None.

Poster

PSTR174. Learning and Memory: Timing and Temporal Processing

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR174.03/SS7

Topic: H.08. Learning and Memory

Support: NIH Grant U19NS104590-01
NIH Grant R01MH122391

Title: Developmentally structured coactivity and plasticity in the hippocampal trisynaptic loop

Authors: *R. HUSZAR[1], M. NARDIN[2], D. HUILGOL[3], J. LIU[5], Z. HUANG[4], C. SAVIN[1], G. BUZSAKI[6];
[1]New York Univ., New York, NY; [2]HHMI Janelia Res. Campus, Ashburn, VA; [4]Duke Univ. Sch. of Med., [3]Duke Univ. Med. Ctr., Durham, NC; [5]New York Univ. Ctr. For Neural Sci., Jersey City, NJ; [6]Neurosci., New York University, Langone Med. Ctr., New York, NY

Abstract: The hippocampus is a key player in learning and memory. Research into this brain structure has long emphasized its plasticity and flexibility (McClelland et al., 1995), though recent reports have come to appreciate its remarkably stable firing patterns (Mizuseki et al., 2013). How plasticity updates networks without destroying their preexisting activity patterns remains an open question, largely due to a lack of experimental access points to neural populations with consistent and known connectivity. Development may provide one such access point (Cossart and Khazipov 2022). We show here that CA1, CA3 and DG principal neurons of the same embryonic birthdate exhibit prominent cofiring across different brain states, including behavior in the form of overlapping place tuning. These features could partially be explained by structured connectivity between locally recorded pyramidal cells and interneurons (Huszár et al., 2022). Prior anatomical work showed that same birthdate excitatory neurons across hippocampal subregions (DG-CA3, CA3-CA1) are synaptically interconnected (Deguchi et al., 2011) displaying highly clustered connectivity on common dendritic branches (Druckmann et al., 2014). This suggests the presence of developmentally installed circuit motifs within and across hippocampal subfields that impose constraints on activity patterns generated in these networks. We explored the consequences of this network connectivity on learning. We birthdated populations of CA3 and CA1 neurons with intrauterine virus injection of AAV9-CAG-Cre followed by adult injection of pAAV-EF1a-DIO hChR2 into CA3 and CA1. After recovery, we implanted high-density silicon probes equipped with optic fibers in ipsilateral CA1 and CA3. Birthdate-defined populations were identified with short-pulse optogenetics in each subfield. Animals were trained in a ‘cheeseboard’ spatial memory task that required encoding novel reward locations on each day (Dupret et al., 2010). At the single cell level, same birthdate neurons remapped no differently than the population in their respective subregion. However, pairs of same birthdate CA1-CA1, CA3-CA3 and CA3-CA1 neurons were more likely to remap together. Excess correlations (i.e. beyond place field similarity and oscillatory modulation; Nardin et al., 2021) among same birthdate pairs rearranged in a distinct manner within and across subfields. This suggests a statistical fingerprint of structured plasticity in interconnected populations, with potential to constrain network models of hippocampal plasticity.

Disclosures: **R. Huszar:** None. **M. Nardin:** None. **D. Huilgol:** None. **J. Liu:** None. **Z. Huang:** None. **C. Savin:** None. **G. Buzsaki:** None.

Poster

PSTR174. Learning and Memory: Timing and Temporal Processing

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR174.04/SS8

Topic: H.08. Learning and Memory

Support: DFG NI 2057/1-1
NIH Grant MH107396
U19 NS107616
U19 NS104590
SFSF Mobility fellowship P500PB_214401

Title: Novelty deteriorates the stability of familiar neural representations

Authors: *N. NITZAN[1], G. BUZSAKI[2];
[1]New York Univ., New York, NY; [2]Neurosci., New York University, Langone Med. Ctr., New York, NY

Abstract: The survival of the organism in a dynamic environment depends on its ability to register novel sensory stimuli. The powerful impact of novelty on neuronal dynamics has been well documented. However, how the neural representations of novelty interact with existing representations of familiar stimuli is not clear. To this end, we analyzed data from mice performing a visual go/no-go change detection task in which subjects are shown a continuous series of natural images and are rewarded with water drops for correctly reporting a *change* in the identity of the image. Each mouse underwent two recording sessions: On the first day, subjects were shown a familiar image set, to which they were exposed during the preceding two-week long training. On the second day, mice were shown a different image set comprising six novel images and two familiar images from the previous set. Mice performed similarly on day 1 when presented with familiar images and on day 2 when the identity of the changed image was novel. Surprisingly, we found that the performance of mice drastically decreased on day 2, when the identity of the changed image was familiar. To examine the neuronal mechanisms of the decreased performance when familiar and novel images are interleaved, we analyzed spiking activity from visual cortical areas and additional subcortical areas. Confirming previous results, we found that novel stimuli on day 2 modulated the activity of a larger fraction of neurons compared to familiar stimuli. However, when interleaved with novel images, both the fraction of cells modulated by familiar images, as well as the magnitude of their modulation decreased in response to the presentation of the familiar images. At the population level, we found that when presented with only familiar stimuli, the visual representations of the different images tended to be embedded at a similar distance from one another, with differences likely owing to stimulus features. In contrast, when interleaved with novel images, we found that familiar images were embedded substantially remotely from novel images, particularly on miss trials, resulting in significant correlations between task performance and neuronal embedding. This observation suggests that successful task performance may depend on the distance between successive neuronal representations. Lastly, it has been shown that the visual cortex exhibits representational drift over the course of minutes. We found that the representations of familiar images drift at a faster rate than novel images, even when familiar and novel stimuli are interleaved, suggesting that the processes underlying the stabilization of neuronal representation are stimulus specific.

Disclosures: **N. Nitzan:** None. **G. Buzsaki:** None.

Poster

PSTR174. Learning and Memory: Timing and Temporal Processing

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR174.05/SS9

Topic: H.08. Learning and Memory

Support: NIH Grant R01MH122391
NIH Grant U19NS107616
NSF Grant 1707316

Title: Cooperative hippocampal and medial prefrontal activity supports memory retrieval but not maintenance: optogenetic probing

Authors: *T. DOHI[1], I. ZUTSHI[2], G. BUZSAKI[3];
[2]New York Univ. SOM, [1]New York Univ., New York, NY; [3]New York Univ., New York University, Langone Med. Ctr., New York, NY

Abstract: Working memory involves the ability to retain the trace of a cue or a spatial location within the brain to guide future decisions. The hippocampal area and medial prefrontal cortex (mPFC) are two regions considered to be critical for working memory. A most prominent view is that the memory of a trace is maintained as persistent activity in the firing of neurons, either within, or coordinated across these brain regions during the delay period.
To test if hippocampal and mPFC activity is required specifically during the delay, we implemented a 'delayed cue-to-place task' where mice were trained on a T-maze. Mice initiated trials by nose-poking at a door at the home base, which triggered a brief (varying duration) visual cue. Following a 1-s long delay, the door opened, and mice ran the length of the track (~1 m) to turn toward the side of the arm where the cue was presented. The setup of the task therefore involves a forced delay (behind the door, 1-s long), and a delay that arises as the mice run down the track (~3-s long).
We trained Parvalbumin (PV)-cre x ChR2 mice to reach high performance levels (70-80%) on this task. Bilateral optic fibers were implanted in the mPFC and the dorsal hippocampus to optogenetically silence these regions. Stimulation duration and timing was targeted to 1 of 6 phases of the trial - (1) cue presentation, (2) delay, (3) cue presentation + delay, (4) first 2 seconds of the run to the choice, (5) last 1.5 seconds of the run, and (6) delay + run.
Unilateral optogenetic silencing of mPFC or hippocampus did not impair behavior at any phase of the task. Bilateral silencing of the hippocampus or the mPFC led to chance-level performance, but only when the silencing was performed during the first 2 seconds of the run to the choice or during the delay + run. All other manipulations, including those targeted to the 1-s delay had no effect on behavior. Extending the delay to 3s before door opening and silencing during those 3 seconds also led to no impairment in performance. Next, we silenced mPFC and hippocampus together but in the opposite hemispheres. Targeting opposite hemispheres during the first 2 seconds of the run to the choice again led to chance performance.
Together, these results suggest that cooperative intra-hemispheric mPFC-hippocampal activity is required specifically during memory retrieval as the mouse is executing its future choice during the central arm, but, surprisingly, not during the initial delay. This outcome suggests that mPFC

systems become indispensable only when retaining information for longer intervals is required. The findings also indicate that an intact single hemisphere is sufficient to maintain working memory, at least in our simple task.

Disclosures: T. Dohi: None. **I. Zutshi:** None. **G. Buzsaki:** None.

Poster

PSTR174. Learning and Memory: Timing and Temporal Processing

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR174.06/SS10

Topic: H.08. Learning and Memory

Title: Within-session representational drift in hippocampal CA1 populations

Authors: *Z. ZHENG[1], A. H. WILLIAMS[2,3], G. BUZSAKI[4];
[1]New York Univ. Ctr. For Neural Sci., New York, NY; [2]New York Univ., New York, NY; [3]Flatiron Inst., New York, NY; [4]New York University, Langone Med. Ctr., New York, NY

Abstract: The balance between rigidity and flexibility is a key function that allows the brain to maintain old information and incorporate new ones. While studies on the flexibility of the hippocampal neural representations focus mainly on the changes induced by novelty in the environment ("remapping") or across a long time span ("representational drift"), it is unclear whether the spatial tunings of place cells change rapidly within a familiar environment within a session. If they do, are the changes coordinated across cells? Are these changes consolidated during sleep? To examine changes in the tuning of CA1 place cells that are sustained across trials, we leverage a change-point detection algorithm and apply a principled statistical framework to a large dataset of simultaneous extracellular recordings. We find a large fraction of place fields spontaneously "switch" between high and low firing rate states. These switches in firing rate persist for multiple trials and can occur anywhere on the track and on any trial, although with a somewhat higher likelihood on early trials. The switching cannot solely be explained by non-stereotypical behavior, like head scanning or multiple physiological parameters that we explored. More pairs of place fields co-switch on the same trial than in a null model assuming independence, suggesting that co-switching is coordinated among sub-populations. Moreover, the co-switching fields show higher coactivation than non-co-switching pairs during post-experience sleep, but not during pre-experience sleep. This result highlights a contrast with the previous work that the overlap of place fields can be predicted by their coactivation during pre-experience sleep. Taken together, place cell activities can be decomposed into a rigid part where pre-existing dynamics constrain place field expressions and a flexible add-on that incorporates experiential changes through co-switching.

Disclosures: Z. Zheng: None. **A.H. Williams:** None. **G. Buzsaki:** None.

Poster

PSTR174. Learning and Memory: Timing and Temporal Processing

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR174.07/SS11

Topic: H.08. Learning and Memory

Support: DFG Walter Benjamin fellowship (grant no. MA 10301/1-1, A.M)
FACES (M.S. and A.M.)
NRSA 5TL1TR001447-07 (M.S.)
NIH Grants MH122391 (G.B.)
U19 NS107616 (G.B.)

Title: Neuronal Modulation by Interictal Epileptiform Discharges: Insights from Intracranial Recordings in Epilepsy Patients and a Transgenic Mouse Model of Alzheimer's Disease

Authors: *A. MASLAROVA[1], M. SOULA[1], J. SHIN[2], A. LIU[2,3,4,1], G. BUZSAKI[5,1];
[1]New York University, Neurosci. Inst., New York, NY; [2]New York Univ. Sch. of Med., New York, NY; [3]New York Univ. Comprehensive Epilepsy Ctr., New York, NY; [4]New York University, Ctr. for Cognitive Neurol., New York, NY; [5]New York University, Langone Med. Ctr., New York, NY

Abstract: IInterictal epileptiform discharges (IEDs) are transient abnormal electrophysiological events that have been linked to poor memory performance in epilepsy and have been observed in patients and rodents with Alzheimer's disease (AD). IEDs interfere with hippocampal sharp-wave ripples (SPW-Rs) and cortical spindles, therefore disrupting hippocampal-cortical communication. While IEDs were traditionally believed to be directly linked to epilepsy, an alternative hypothesis is that they form a continuum with hippocampal ripples in an imbalanced network in a broad spectrum of neurodegenerative conditions. To address this hypothesis, we compared the firing modulation of hippocampal neurons by IEDs in two distinct settings. First, we investigated hippocampal and parahippocampal IEDs in epilepsy patients implanted with combined micro/macrowire intracranial EEG electrodes for surgical resection planning. While all patients suffered from drug-resistant epilepsy, some of them had an extratemporal focus without hippocampal involvement. Next, to assess specific neuronal modulation in the CA1 region and the interference with SPW-Rs, we additionally analyzed IEDs in high-density hippocampal recordings from adult AD transgenic mice (APP/S1), carrying two human transgenes associated with AD. We observed IEDs in epilepsy patients with temporal and frontal lobe foci and in AD mice, independent of seizure generation. The IEDs showed similar local field potential features and waveforms in both subject groups, along with comparable modulation of pyramidal neurons and interneurons. IEDs generally recruited pyramidal neurons into firing and silenced interneurons. In addition, IEDs suppressed the incidence and altered the properties of physiological SPW-Rs. These results suggest that IEDs may represent a single-standing pattern of activity similar across species and neurodegenerative diseases. So far, treatment in both epilepsy and AD have focused on suppressing seizures with antiepileptic drugs. Nevertheless, patients with cognitive deficits may benefit from selective treatment strategies for IEDs.

Disclosures: **A. Maslarova:** None. **M. Soula:** None. **J. Shin:** None. **A. Liu:** None. **G. Buzsaki:** None.

Poster

PSTR174. Learning and Memory: Timing and Temporal Processing

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR174.08/SS12

Topic: H.08. Learning and Memory

Support: MH122391
U19NS107616
R90DA043849

Title: Computational model of the role of disinhibition in hippocampal CA1 activity

Authors: *L. GREEN[1], G. BUZSAKI[2], J. RINZEL[3];
[1]NYU Sch. of Med., New York, NY; [2]New York University, Langone Med. Ctr., New York, NY; [3]New York Univ. Ctr. for Neural Sci., New York, NY

Abstract: Recent work suggests disinhibition plays a role in shaping place cell activity in the hippocampal CA1 region. From a computational perspective, disinhibition provides a mechanism for self-sustained activity, suggesting attractor-like activity in CA1 without the need for recurrent excitation. Most previous studies on attractor dynamics utilize recurrent excitation, as is the case in the CA3 region. In turn, it is tacitly assumed that the target CA1 region functions simply as a relay or output node, without exploiting the computational properties of CA1 circuitry. In a reduced firing rate model, we confirm that disinhibition coupled with broad lateral inhibition can lead to attractor dynamics. That is, with spatially uniform and strong enough input to excitatory and inhibitory cells the distributed rate model is capable of a spatially-localized ‘bump’ of activity. We then develop a spiking population model of CA1 place cell activity based on the same circuitry. We model place cell activity along a linear track in response to spatiotemporal input. Compared to all-to-all circuitry, the model with disinhibition offers improved signal-to-noise and more stable maintenance of temporal organization of place cell activity in response to changes in input. This observation is consistent with known functions of attractor models and proposed roles of the hippocampus, to compress information while maintaining encodability.

Disclosures: **L. Green:** None. **G. Buzsaki:** None. **J. Rinzel:** None.

Poster

PSTR174. Learning and Memory: Timing and Temporal Processing

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR174.09/SS13

Topic: H.08. Learning and Memory

Title: Distinct circuit-level operations of parvalbumin and somatostatin expressing interneurons in hippocampal CA3 and dentate gyrus

Authors: A. E. HEYNOLD[1], ***T. HAINMUELLER**[2], G. BUZSAKI[1];
[1]Neurosci. Inst., New York University, Langone Med. Ctr., New York, NY; [2]Psychiatry, New York Univ., New York, NY

Abstract: The hippocampus is essential for the storage and recall of declarative memories. It comprises a number of different anatomical subfields that are connected predominantly in a feed-forward loop starting with the dentate gyrus and progressing through the CA2/3 and 1 areas. Each of these subregions makes unique contributions to hippocampal circuit- and memory-related functions (Buzsaki 1989, *Neuroscience,* Hainmueller and Bartos, 2020, *Nat. Rev Neurosci.*). In addition to the specialized principal cells in the respective hippocampal subfields, different types of GABAergic interneurons help to regulate and coordinate hippocampal networks (Roux and Buzsaki, 2015, *Neuropharmacology*). While there is a solid body of work describing activities and functions of interneurons in hippocampal CA1, less is known about potentially unique roles of inhibitory circuits in 'deeper' hippocampal subfields such as CA3 or the dentate gyrus which are the first to process new input from the entorhinal cortex.
Here we aim to close this gap by selective optogenetic labeling and manipulations of different genetically defined subpopulations of interneurons in these hippocampal regions. We describe basic electrophysiological characteristics of parvalbumin and somatostatin expressing interneurons in the dentate gyrus and CA3 and study the impact of their activation and silencing on the dynamics of their surrounding principal cell networks. Our results indicate that different types of interneurons play unique and subfield-specific roles in regulating population activity motives and may thereby make discernible contributions to the cognitive operations supported by the respective networks.

Disclosures: **A.E. Heynold:** None. **T. Hainmueller:** None. **G. Buzsaki:** None.

Poster

PSTR174. Learning and Memory: Timing and Temporal Processing

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR174.10/SS14

Topic: H.08. Learning and Memory

Support: 1U19NS107616-01

Title: Neuromodulation of hippocampal networks by acetylcholine and oxytocin, interaction via the lateral septum

Authors: *Y. ZHANG[1], M. KARADAS[2], J. LIU[3], Y. LI[4], R. W. TSIEN[5], G. BUZSAKI[6]; [1]nyu grossman school of medicine, NEW YORK, NY; [2]NYU Langone Hlth., New York, NY; [3]New York Univ., new york, NY; [4]Peking Univ., Peking Univ., Beijing, China; [5]NYU Grossman Sch. of Med., NYU Grossman Sch. of Med., New York, NY; [6]Neurosci., New York University, Langone Med. Ctr., New York, NY

Abstract: One of the main postulated roles of subcortical neuromodulators is to control brain states. How the different neuromodulators compete and cooperate at the temporal scales of finer network changes have remained a major question. We investigated how fluctuation of Ach and OXT levels correlate with brain state changes and interact with each other, using G-protein-coupled receptor activation-based acetylcholine ($GRAB_{ACh}3.0$, $GRAB_{rACh}1.4$) and oxytocin ($GRAB_{OXT}1.7$) sensors with a fiber-photometric fluorescence readout in mice. Concurrently, we recorded hippocampal network activity using high-density electrode arrays from multiple regions and layers of the hippocampus, while also capturing the animals' movements during spontaneous behaviors. While Ach and OXT worked in parallel during NREM packets, transitions from NREM to REM were characterized by a surge of Ach and a further decrease of OXT. High Ach was correlated with population synchrony and gamma oscillations during active waking but sharp wave ripples (SPW-R) of immobility and NREM were associated with minima of Ach signal. Optogenetic control of Ach and OXT neuron confirmed the active role of these neuromodulators in the observed correlations. Activation of medial septal cholinergic neurons induced a rapid phasic (<2s) and sustained decrease of OXT in the hippocampus, mimicking the lag between Ach and OXT during spontaneous behaviors. Synchronous activity of hippocampal population consistently reduced OXT activity, and we show that this effect is mediated by the lateral septum-hypothalamus inhibitory path. Our findings demonstrate how cooperative actions of Ach and OXT neuromodulators allow target circuits to perform specific functions.

Disclosures: **Y. Zhang:** None. **M. Karadas:** None. **J. liu:** None. **Y. Li:** None. **R.W. Tsien:** None. **G. Buzsaki:** None.

Poster

PSTR174. Learning and Memory: Timing and Temporal Processing

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR174.11/SS15

Topic: H.08. Learning and Memory

Support: R01MH122391
U19NS104590
U19NS107616

Title: The neural mechanism of learning to generalize

Authors: *K. KISELEV[1], W. YANG[2,3], G. BUZSAKI[4];
[1]New York Univ. Ctr. For Neural Sci., New York, NY; [2]Ctr. for Neural Sci., New York Univ., New York, NY; [4]Neurosci., [3]New York University, Langone Med. Ctr., New York, NY

Abstract: The ability to generalize across diverse contexts and use generalizable knowledge to act in new situations is a remarkable feat of the brain. Yet the neural mechanism of the learning process of generalization is unknown. We designed a behavioral task where mice learn a cue-guided navigation task. Mice first learned rules in a familiar environment over 14 sessions and then generalized this experience to 2 distinct novel contexts. Our behavioral results revealed a unique 'learning to generalize' phenomenon where once mice learned to generalize information in the first novel maze (over the course of 4 sessions), they displayed 'one shot learning' in the next session where they rapidly generalized to the second novel contexts within a single session. While most past studies characterize the representation in the brain after learning, we record from the hippocampus during learning as well as sleep. We are in the process of identifying and further dissecting neuronal mechanisms in the hippocampus that relate to the consolidation of specific experiences and generalization of experiences.

Disclosures: K. Kiselev: None. **W. Yang:** None. **G. Buzsaki:** None.

Poster

PSTR174. Learning and Memory: Timing and Temporal Processing

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR174.12/SS16

Topic: H.08. Learning and Memory

Support: EU Horizon 2020 Marie Skłodowska-Curie grant agreement No 892957

Title: Functional coupling of dorsal midline thalamic neurons to the prelimbic cortex and the ventral subiculum

Authors: *G. KOMLÓSI, G. BUZSAKI;
Neurosci. Inst., NYU Langone Grossman Sch. of Med., New York, NY

Abstract: The dorsal midline thalamus (dMT) is a group of non-sensory thalamic nuclei that has been implicated in arousal and in the homeostatic maintenance of sleep choreography. Neocortical slow oscillations and hippocampal sharp wave ripples (SPW-R) are the hallmarks of non-rapid eye-movement (NREM) sleep and reflect synchronous activation of neocortical and hippocampal neuronal ensembles, respectively. It is well established that thalamic activity in general is tightly and uniformly coupled to the cortical slow oscillations alternating between active UP and silent DOWN states. It was recently shown that neuronal activity in the dMT is also coupled to the cortical slow oscillation and can influence UP and DOWN state transitions. In addition, the anterior but not the posterior part of the dMT receives a strong innervation from the ventral subiculum, the main output of the hippocampus. However, there is very little known about the interaction between dMT and hippocampal activity during sleep. Since different parts

of the dMT are differentially connected to the neocortex and the hippocampal formation we hypothetized that dMT neurons respond heterogenously during sleep. We performed large-scale unit recordings from dMT neurons during unrestrained sleep-wake behavior, while simultaneously recorded population activity from the prelimbic cortex and ventral hippocampus/subiculum. We found that neurons are differentially coupled to cortical DOWN states and hippocampal SPW-Rs in the posterior and anterior segments of dMT, and they show different firing characteristics during sleep-wake behavior. Surprisingly, we found that a fraction of dMT neurons are selectively active during cortical DOWN states and their activity is either negatively or not correlated with hippocampal SPW-Rs. When the activity of these DOWN state active neurons (DSA) are compared with the rest of the population, we found that DSA neurons showed higher correlation with each other than with with non-DSA neurons in both in NREM sleep and during wakefulness, suggesting the presence of two antagonistic subnetworks within the dMT. These findings demonstrate how neocortical and hippocampal/subicular regions can influence this “limbic” thalamic nucleus. In the reverse direction, dMT may coordinate activity across wide cortical and subcortical areas.

Disclosures: G. Komlósi: None. **G. Buzsaki:** None.

Poster

PSTR174. Learning and Memory: Timing and Temporal Processing

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR174.13/SS17

Topic: H.08. Learning and Memory

Support: R01MH122391
U19NS104590
U19NS107616

Title: Credit assignment and memory tagging: selection of experience for consolidation by hippocampal sharp wave ripples

Authors: *W. YANG[1], C. SUN[2], R. HUSZAR[1], G. BUZSAKI[3];
[1]New York Univ., New York, NY; [2]MILA, Montreal, QC, Canada; [3]New York University, Langone Med. Ctr., New York, NY

Abstract: We do not remember every moment in life. How does the brain select which aspects of the experience are worth retaining for long-term consolidation and future use?
Hippocampal sharp wave ripples (SPW-Rs) have been demonstrated to support memory consolidation during sleep. While previous works suggest the important role of SPW-Rs for memory consolidation, the memory selection mechanism remains unknown.
Using UMAP, we observed that population activity of hippocampal neurons varies systematically across trials. We observed that SPW-Rs did not occur on every trial but instead tended to concentrate on certain trials. The trials after which SPW-R occurred during maze

learning varied across sessions and across animals. When brain state changed during reward consumption and sharp wave ripples (SPW-Rs) occurred on some trials, their unique spike content corresponded to the preceding maze traversal. Importantly, during post-experience sleep in the home cage, SPW-Rs replay spike content of those trials that were tagged by waking SPW-Rs. This relationship was not observed between awake SPW-Rs and SPW-Rs of pre-experience sleep.
Our results suggest that during learning, awake SPW-Rs 'tag' particular memory content, which are then replayed selectively during post-experience sleep. We hypothesize that SPW-Rs provide a post-hoc editing mechanism to select which aspects of experience are preserved for future use.

Disclosures: **W. Yang:** None. **C. Sun:** None. **R. Huszar:** None. **G. Buzsaki:** None.

Poster

PSTR174. Learning and Memory: Timing and Temporal Processing

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR174.14/SS18

Topic: H.08. Learning and Memory

Support: NIH Grant 1R01NS113782-01A1
NIH Grant 2TL1TR001447-06A1

Title: Transcranial Radio-Frequency Stimulation (TRFS) as a non-invasive neuromodulation technique

Authors: *O. YAGHMAZADEH[1], G. BUZSAKI[2];
[1]New York Univ. Neurosci. Inst., New York, NY; [2]New York Univ., New York University, Langone Med. Ctr., New York, NY

Abstract: Non-invasive brain stimulation techniques provide an unprecedented opportunity to probe and modify the brain in health and disease. Some of established such methods, e.g. transcranial electrical stimulation, transcranial magnetic stimulation, and transcranial focused ultrasound stimulation, have led to numerous novel insights into brain function and are now widely used in clinical practice, including rehabilitation and treatment of mental disease. Each of these methods has proven some benefits but also comes with limitations. Therefore, the search for novel approaches to non-invasively modulate neural activity is of great interest to the scientific community, providing a wider range of possible treatments for future clinical applications. Here, we report the application of radio-frequency (RF) energy radiation for non-invasive modulation of ongoing neural activity in-vivo in mice. Using in-house developed RF circuits, we have been able to induce controlled and safe temperature rises in brains of rodents (with hemispheral preference) without affecting the body temperature. It is well established that temperature changes induce modulation of ongoing neural activity. Our experiments using functional Magnetic Resonance Imaging (fMRI) and Ca^{2+} fiber-photometry, show that our RF stimulation with thermal effects on the brain can induce significant changes in neural activity and

brain physiology in mice. We report both suppression and excitation of neural activity in TRFS-exposed mice brains in-vivo. These preliminary experiments open new horizons in adding a novel technique to the arsenal of non-invasive brain stimulation paradigms with potential benefits in the treatment of brain disorders.

Disclosures: O. Yaghmazadeh: None. **G. Buzsaki:** None.

Poster

PSTR174. Learning and Memory: Timing and Temporal Processing

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR174.15/SS19

Topic: H.08. Learning and Memory

Support: NIH R01 NS116357
NIH RF1 AG072497
CURE Taking Flight Award
NIH F32 NS116416

Title: Inhibitory theta phase locking in the healthy and epileptic hippocampus and its impact on seizures and cognition

Authors: *Z. CHRISTENSON WICK[1], P. A. PHILIPSBERG[1], S. I. LAMSIFER[2], C. KOHLER[1], E. KATANOV[1], Y. FENG[1], D. J. CAI[1], T. SHUMAN[1];
[2]Nash Family Dept. of Neurosci., [1]Icahn Sch. of Med. at Mount Sinai, New York, NY

Abstract: Network-wide oscillations, such as theta, orchestrate and organize the spiking of individual neurons in a phenomenon known as phase locking. Phase locking has long been thought to maintain excitatory-inhibitory homeostasis and coordinate cognitive processes. We've recently found altered theta phase locking of inhibitory neurons in the dentate gyrus of epileptic mice with spontaneous seizures and cognitive deficits. While phase locking has been widely studied in a variety of contexts using correlational methods, the direct, causal influence of this phenomenon has never been determined. Thus, we aimed to directly test the hypothesis that inhibitory theta phase locking can bidirectionally control seizures and cognitive performance in control and epileptic mice. To test these hypotheses, we developed a low-latency closed-loop optogenetic system to bidirectionally control inhibitory phase locking to theta in head-fixed control and pilocarpine-treated epileptic mice navigating a virtual track. Using opto-tagging strategies, we first identified the preferred firing phase of parvalbumin (PV)+ and somatostatin (SOM)+ dentate interneurons in control and epileptic mice. We then applied our closed-loop system to lock the spiking of these dentate interneurons to their preferred or non-preferred phase of theta while measuring seizure activity and accuracy while navigating a virtual environment.Using our closed-loop optogenetic system in awake behaving mice, we have validated our ability to precisely alter the phase locking of hippocampal interneurons. Using this system, we have found that mis-aligning inhibitory spiking to the peak of theta increases seizure

susceptibility in otherwise healthy, control mice. Furthermore, in epileptic mice, re-aligning inhibitory spiking to the trough of theta diminishes pathological epileptic activity compared to stimulating at the peak of theta. Finally, we have preliminary data demonstrating that precise theta phase locking of dentate gyrus inhibitory neurons influences performance on a demanding dentate-dependent virtual navigation task.Together, these data suggest that theta phase locking of inhibitory spiking plays an important and causal role in two of the most concerning elements of epilepsy: hyperexcitability and cognitive deficits. Gaining deeper insights into the impacts of inhibitory theta phase locking may reveal the potential of oscillation-driven stimulation as an effective epilepsy therapeutic.

Disclosures: Z. Christenson Wick: None. **P.A. Philipsberg:** None. **S.I. Lamsifer:** None. **C. Kohler:** None. **E. Katanov:** None. **Y. Feng:** None. **D.J. Cai:** None. **T. Shuman:** None.

Poster

PSTR174. Learning and Memory: Timing and Temporal Processing

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR174.16/SS20

Topic: H.08. Learning and Memory

Support: CURE Taking Flight Award
NIH Grant R01 NS116357
NIH Grant RF1 AG072497
NIH Grant F32 NS116416

Title: Phaser: an open-source tool for real-time, phase-targeted manipulations during endogenous oscillations

Authors: *P. A. PHILIPSBERG[1], Z. CHRISTENSON WICK[3], S. I. LAMSIFER[2], C. KOHLER[1,4], E. KATANOV[1,5], Y. FENG[6], C. HUMPHREY[1,5], T. SHUMAN[6];
[2]Nash Family Dept. of Neurosci., [1]Mount Sinai Sch. of Med., New York, NY; [3]Icahn Sch. of Med. at Mount Sinai, New York, NY; [4]New York Univ., New York, NY; [5]Hunter College, CUNY, New York, NY; [6]Icahn Sch. of Med. At Mount Sinai, New York, NY

Abstract: The coordination of neural activity relative to ongoing oscillations, (e.g., in the form of spike-phase coupling, phase-phase coupling, and phase-amplitude coupling), has long been hypothesized to play a crucial role in both facilitating the flow of information in cognitive processing, and in maintaining excitatory-inhibitory balance. Causal investigations into these relationships, however, have thus far been limited by the significant technical challenges associated with specifically and precisely manipulating activity relative to endogenous oscillations. In order to address this, we have developed a closed-loop optogenetic system for cell-type specific stimulation phase-locked to endogenous oscillations. Our closed-loop system, PhaSER (Phase-locked Stimulation to Endogenous Rhythms), uses low-latency signal processing and auto-regressive forward prediction to enable real-time phase estimation and temporally

precise manipulations. Support for two independent stimulation channels enables bidirectional modulation of neuronal activity through the use of the BiPOLES construct. Additionally, multiple stimulations patterns are supported, including square wave pulse trains and sinusoidal modulation. Here, we evaluate the phase-targeting performance of this tool and test its ability to manipulate the phase-locking properties of somatostatin-expressing (SOM+) interneurons relative to hippocampal theta oscillations. We show that PhaSER is able to accurately target specific phases of theta in real-time across a range of physiological theta powers in awake, behaving mice. Additionally, we show that phase-locked stimulations are able to shift the preferred firing phase of hippocampal SOM+ interneurons without altering the referenced theta power or phase. PhaSER is highly flexible and can be used to target cell-type specific manipulations to any phase of an endogenous oscillation. Phase specific manipulations will be crucial for discovering the causal contributions of altered neural synchrony, which is seen across many neurological disorders including epilepsy, Alzheimer's disease, and autism spectrum disorders. We have released PhaSER as an open-source tool to facilitate its use in future research.

Disclosures: **P.A. Philipsberg:** None. **Z. Christenson Wick:** None. **S.I. Lamsifer:** None. **C. Kohler:** None. **E. Katanov:** None. **Y. Feng:** None. **C. Humphrey:** None. **T. Shuman:** None.

Poster

PSTR175. Human Navigation: Neurophysiology, Cognitive Mechanisms, Deficits

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR175.01/SS21

Topic: H.09. Spatial Navigation

Support: NIH Grant R01-MH104606

Title: Human single-neuron correlates of spatial navigation and memory performance

Authors: *C. Z. HAN[1], T. DONOGHUE[1], L. KUNZ[2], J. JACOBS[1];
[1]Biomed. Engin., Columbia Univ., New York, NY; [2]Univ. of Bonn, Bonn, Germany

Abstract: A multitude of investigations have explored the neural basis of spatial navigation and memory-related processes represented in hippocampus and surrounding structures in the medial temporal lobe (MTL). Using naturalistic tasks that involve complex human behaviors has become an established approach to characterize neural activity when encoding information during spatial navigation of complex environments. A large body of the existing literature demonstrates that the activity of specific cell types can show selective spatial modulation (e.g. place cells) in task-relevant ways. While previous research has demonstrated these kinds of single neuron activity patterns, for example by analyzing navigation activity during encoding periods, what has been less examined is the precise relationships between such neural activity and behavioral performance, including trial-by-trial performance measures and analyzing activity during recall periods. In order to address these questions, we recorded neural data from the

hippocampus and surrounding areas in the MTL of human neurosurgical patients with implanted microwires while they performed a spatial episodic memory task named Treasure Hunt. In each trial of Treasure Hunt, participants used a joystick to navigate an open arena on a virtual beach and encountered treasure chests that contained items. Participants were instructed to remember the location of the presented items, so that they can later report the location of each encountered item. Examining the navigation periods, we first analyzed single-neuron activity for task-related modulations during encoding which has been shown in our previous work, including spatial target cells whose responses reflect the position of presented items, and serial position cells that encode the sequential order of encountered items. Next, we examined and found relationships between single neuron activity and behavioral performance by correlating the normalized firing rate of each cell to the normalized distance error of recall responses. In addition, by quantifying the similarity between the movement during recall and encoding behavior during navigation, we found an association between the patterns of recall responses and single neuron activity in the MTL. This allowed us to examine participants' navigation and recall strategies, connecting them to underlying neural activity as a predictor of memory strength. Altogether, these results demonstrate the benefits of connecting continuous behavioral measures to neural activity in order to specify the relationship between neural activity during encoding and behavioral performance during recall.

Disclosures: C.Z. Han: None. **T. Donoghue:** None. **L. Kunz:** None. **J. Jacobs:** None.

Poster

PSTR175. Human Navigation: Neurophysiology, Cognitive Mechanisms, Deficits

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR175.02/SS22

Topic: H.09. Spatial Navigation

Title: High and low theta oscillations in human prefrontal cortex are traveling waves

Authors: E. ZABEH[1], ***S. KANG**[1], M. TSITSIKLIS[1], E. H. SMITH[2], G. M. MCKHANN II[3], C. A. SCHEVON[3], S. SHET[4], J. JACOBS[1];
[1]Columbia Univ., New York, NY; [2]Neurosurg., Univ. of Utah, Salt Lake City, UT; [3]Columbia Univ. Med. Ctr., New York, NY; [4]Baylor Col. of Med., Houston, TX

Abstract: The hippocampal theta oscillation, a rhythmic oscillation in the range of ~4-10 Hz, plays a crucial role in memory formation and spatial navigation. Recent studies have revealed that this oscillation in the human brain is rather complex, functionally segregated into two distinct regimes of high-frequency and low-frequency oscillations, and that it also behaves as a traveling wave (TW). In this study, we traced the spatial structure between cortical neuronal activity and theta oscillations in humans. We recorded simultaneous field potentials and neuronal spiking activity in prefrontal cortex (PFC) of patient undergoing intracranial monitoring for epilepsy engaging in a memory-navigation task, using multi-electrode "Utah" arrays. Our analysis revealed the presence of oscillations in the human PFC in both the high and low portions

of the theta band. We further examined the spatial structure of this signal by analyzing spatio-temporal organization of these oscillations across the recording grid. We verified each of high and low theta oscillations distinctively exhibit characteristics of TWs. By analyzing the physical properties of these TWs and their interaction with neuronal spiking, we aim to shed light on how oscillations modulate the spatial structure of cortical processing across different frequency bands.

Disclosures: **E. Zabeh:** None. **S. Kang:** None. **M. Tsitsiklis:** None. **E.H. Smith:** None. **G.M. McKhann II:** None. **C.A. Schevon:** None. **S. Shet:** None. **J. Jacobs:** None.

Poster

PSTR175. Human Navigation: Neurophysiology, Cognitive Mechanisms, Deficits

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR175.03/SS23

Topic: H.09. Spatial Navigation

Support: NIH Grant R01-MH104606

Title: Variability across methods in the identification and characterization of place cells in human data

Authors: ***W. ZHANG**[1], T. DONOGHUE[1], S. E. QASIM[2], J. JACOBS[1];
[1]Biomed. Engin., Columbia Univ., New York, NY; [2]Psychiatry, Mount Sinai Sch. of Med., New York, NY

Abstract: Spatial navigation is a complex cognitive process enabling humans and animals to determine routes towards goals, navigate the environment, and make movement decisions. At the core of this process are hippocampal place cells, neurons that exhibit selective firing patterns at specific locations in the environment. However, the choice of detection method results in variations in the population of identified place cells, and subsequently leads to variations in measured features such as spatial selectivity, firing fields and firing rate. Understanding this variability is important for interpreting and comparing findings across studies, as it highlights the influence of methodological choices on our understanding of place cell properties. While previous research in animal models has highlighted the influence of different place-cell detection methods, it is also important to examine this question in human data, given it's idiosyncrasies and unique characteristics. This study therefore aims to examine the differences in detection methods, including spatial information and ANOVA methods. The study also seeks to understand how these methodological variations impact the identified place cell populations, and compare features such as spatial selectivity, firing fields, and firing rates. To do so, we analyzed data recorded from human neuro-surgical patients who have implanted electrodes for clinical reasons, including microwires which capture single-unit activity. For this investigation, patients participated in a computer-based virtual navigation spatial task, in which they navigated along a linear track and were instructed to learn and recall the location of presented items, while recording single micro-wires in the medial temporal lobe (MTL). We found that the spatial

information method identified a distinct set of place cells compared to the ANOVA method, indicating that these two detection methods capture different populations of place cells with limited overlap. The study further characterized these identified place cells by analyzing their peak firing rates and place fields, unveiling distinct patterns and characteristics of place cells that were detected with each approach. Overall, by comparing place-cell detection methods we found method dependent variability of place cells in human data, demonstrating the importance of having a clear and consistent criteria for reliable comparisons of place cell properties across experiments in order to better understand the neural substrate of spatial navigation and memory processes.

Disclosures: **W. Zhang:** None. **T. Donoghue:** None. **S.E. Qasim:** None. **J. Jacobs:** None.

Poster

PSTR175. Human Navigation: Neurophysiology, Cognitive Mechanisms, Deficits

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR175.04/SS24

Topic: H.09. Spatial Navigation

Title: Environment schema influences in strategic spatial navigation

Authors: *P. MAXIM, T. I. BROWN;
Sch. of Psychology, Georgia Inst. of Technol., Atlanta, GA

Abstract: Studies on spatial schemas have primarily come from rodent studies examining the development of task representations in the animal's brain. Such studies provide support for accelerated learning of novel spatial associations when prior associations already exist, and notably, there seems to be rapid disengagement of the hippocampus when encoding new experiences within an existing cognitive map. Research has set out to test whether existing spatial knowledge can also benefit novel learning in humans, and if there are similar neural characteristics during prospective planning and recall of memories related to existing spatial knowledge. The present study tested 19 healthy young adult participants (ages 18-28) across two days in a virtual navigation task and used fMRI to examine complementary views of how the medial temporal lobe and medial prefrontal cortex (mPFC) contribute to route planning and navigation. Participants trained on specific paths through six virtual towns, and after 24 hours were tested during fMRI on memory of these now-familiar routes (FR). They were then given novel goals and permitted to navigate to the goal using any strategy (unbound to the FR). Some trials were designed so that the optimal shortcut followed the same direction as the FR (forward-shortcut), while in others the optimal route required traveling backward relative to the FR direction (backward-shortcut) - i.e., greater heading conflict between the optimal and FR. Behavioral analyses show participants follow closer to the optimal route for forward-shortcuts. But during backward-shortcut trials, when given a choice between a) backtracking or shortcutting in a "conflicting" direction with the FR vs. b) traveling along the FR direction, participants show a significant bias toward more closely following the FR than the optimal

shortcut. Univariate and multivariate fMRI analyses revealed: 1) Functional differences in subdivisions of the mPFC, sometimes agreeing, but other times responding differently across various stages of navigation (e.g., planning vs. goal arrival), and differing in how they explain individual differences in navigation behavior. 2) Broad agreement between when and how the hippocampus (right hemisphere in particular) and mPFC (posterior-ventral mPFC in particular) are engaged for task stages, represent environments, and track participant differences – a finding which aligns well with their anatomical interconnections, but may contradict the competitive view from models of schema memory.

Disclosures: P. Maxim: None. **T.I. Brown:** None.

Poster

PSTR175. Human Navigation: Neurophysiology, Cognitive Mechanisms, Deficits

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR175.05/SS25

Topic: H.09. Spatial Navigation

Support: NIH/NIA R01AG073250
The Shurl and Kay Curci Foundation

Title: Environment geometry informs distance error estimation in navigation

Authors: *J. LONG, E. HERRERA, Y. LI, F. OLIVEIRA, R. AHMED, J. DEWAL, T. I. BROWN;
Georgia Inst. of Technol., Atlanta, GA

Abstract: Recent studies have reported contradictory findings about how we encode geometric information during navigation, and rodent research has revealed that the geometric shape of an environment may impact grid cell signaling differently. However, little is known about the relationship between grid cells and geometric information processing in humans. In this study, we aimed to investigate the contradictory findings tying geometry to distance error estimation in environments, with an eye on how training history may be a factor. Participants performed a virtual navigation task from an egocentric perspective in which they learned routes of six object locations in four open-field environments, consisting of two trapezoids and two squares. The presentation order of the environments was randomized as well. Participants were then tested on their spatial memory of the object locations and sequences by navigating the same environments through a first-person perspective once again, and placing the objects in the learned location and order as accurately as they could. In each environment, participants completed three repetitions of route learning and route testing. Participants' distance error estimation significantly decreased between the first and third route tests. Interestingly, participants also performed similarly on average across both sets of square and trapezoid environments - however, training history mattered: performance in the second trapezoid that participants encountered was significantly better than the performance in the second square that participants encountered. These results

support arguments that more-distinctive geometry may be beneficial for learning, even though it distorts grid cells in rodents, but the results also underscore the influence of factors other than environment geometry on distance error estimation in route tracing tasks. These findings inform planned analyses of spatial representations in the human brain with this paradigm, and by uncovering these patterns, we expand on previous geometric navigation research and contribute novel insight into the mechanisms of spatial navigation.

Disclosures: J. Long: None. **E. Herrera:** None. **Y. Li:** None. **F. Oliveira:** None. **R. Ahmed:** None. **J. DeWal:** None. **T.I. Brown:** None.

Poster

PSTR175. Human Navigation: Neurophysiology, Cognitive Mechanisms, Deficits

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR175.06/TT1

Topic: H.09. Spatial Navigation

Support: UK Economic and Social Research Council Grant (ES/R011494/2)

Title: Spatial proximity determines competition between landmarks in open virtual environments

Authors: *E. HERRERA[1], J. M. AUSTEN[2], G. P. URCELAY[2];
[1]Sch. of Psychology, Georgia Inst. of Technol., Atlanta, GA; [2]Univ. of Nottingham, Nottingham, United Kingdom

Abstract: Different studies have documented either competition or its absence between different sources of information (e.g., landmarks and geometry) during navigation, resulting in intense theoretical debates in spatial cognition. However, little is known about which variables determine whether competition is observed. In these experiments, we aimed to replicate in humans, findings in food-storing birds (Clark's nutcrackers), which suggest that landmarks close to the goal location impair learning about distal landmarks, and this is attenuated when assessing the effect of distal landmarks on learning about the proximal ones. Three groups of participants were trained in a virtual open environment featuring orientation cues, and they had to find a hidden goal with reference to four landmarks that were arranged in the shape of a cross. Critically, for different groups the landmarks were placed at different distances from the goal (close, intermediate and far). However, two of the four landmark distances (i.e., placed at 50 and 70 Virtual Units ─VU hereafter─ from the goal) were common across all three groups and a fourth Control Group was trained with these critical 50 and 70 VU landmarks only, to allow a comparison of the extent of competition. Following training, the 50 and 70 VU landmarks were tested, and the distance between where the goal actually was and participants' estimations was assessed. Of interest was how well participants performed when tested with the 50 and 70 VU landmarks, which were common amongst the three experimental and Control Groups. Landmarks near the goal (10, 30 VU) impaired learning about landmarks further away from the goal (50 and 70 VU), but the opposite was not true. A second experiment, in which the number

of training trials was extended from 6 to 16, revealed similar results. Consistent with the results in birds, we observed better performance to landmarks 50 and 70 in the groups with more distal (90, 110 VU) landmarks ─similar to the performance in the Control Group─, suggesting that competition was greater in the groups with closer landmarks. Thus, the extent of competition was dependent on the spatial distance between the landmarks and the goal. These results reveal that proximity is a critical determinant of competition between landmarks in human navigation.

Disclosures: E. Herrera: None. **J.M. Austen:** None. **G.P. Urcelay:** None.

Poster

PSTR175. Human Navigation: Neurophysiology, Cognitive Mechanisms, Deficits

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR175.07/TT2

Topic: H.09. Spatial Navigation

Support: NIH (NIA) 2R01-AG011230

Title: Age-sensitive neural and cognitive correlates of cognitive mapping: Insights into wayfinding deficits

Authors: *A. N. CHARGO[1,2], C. L. DAHLE[2], N. RAZ[3,4], A. M. DAUGHERTY[1,2];
[1]Dept. of Psychology, [2]Inst. of Gerontology, Wayne State Univ., Detroit, MI; [3]Dept. of Psychology, Stony Brook Univ., Stony Brook, NY; [4]Max Planck Inst. for Human Develop., Berlin, Germany

Abstract: Independent living becomes progressively difficult with advanced age, and navigating one's environment is essential for maintaining independence. Wayfinding is key to navigation, and it relies on cognitive maps - mental representations of the environment. Yet, the neural and cognitive correlates of age-related differences in cognitive mapping remain unclear. To address this lacuna in knowledge we examined a community sample of 275 participants (65.8% female), age 18-79 years, who completed a virtual Morris Water Maze, followed by a series of map reproduction tasks. Exploratory two-step cluster analysis using BIC log-likelihood fit (BIC = 737.71) identified 3 cognitive mapping profiles with different combinations of measures (Measure × Mapping Profile: $F_{8, 520} = 66.51$, $p < 0.001$, $\eta_p^2 = 0.51$): recall of the environment map ($p < 0.05$), spatial precision ($p < 0.05$), and navigation efficiency ($p < 0.05$). Discriminant function analysis (DFA) rank-order comparisons revealed that individuals assigned to the poor mapping-poor efficiency cluster were older, had worse immediate spatial association recall, worse immediate and delayed episodic recall, and smaller volumes of prefrontal cortex and striatum as compared to high mapping-high efficiency individuals (all loadings > |0.3|). This collectively accounted for 88.6% of between-group variance ($\chi;;^2 = 77.76$, $df = 32$, $p < 0.001$). Hippocampal volume weakly differentiated cognitive mapping profiles (loading = 0.22) and was predictive only among younger adults (loading = 0.28). Age-stratified DFA revealed a shift to extra-hippocampal regions (hippocampal loading $\leq$ 0.18; prefrontal cortex and striatum loadings

≤ 0.24) and a greater contribution of cognitive correlates including spatial association recall, cognitive processing speed, and episodic recall (all loadings > |0.3|) among middle-age and older adults. Our findings highlight the unique role of neural and cognitive correlates that support cognitive mapping and a shift in these correlates with advanced age. As wayfinding deficits are common in community-dwelling older adults who are at risk for dementia, understanding neural and cognitive sources of decline in cognitive mapping may help in mitigating this risk.

Disclosures: **A.N. Chargo:** None. **C.L. Dahle:** None. **N. Raz:** None. **A.M. Daugherty:** None.

Poster

PSTR175. Human Navigation: Neurophysiology, Cognitive Mechanisms, Deficits

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR175.08/TT3

Topic: H.09. Spatial Navigation

Title: Impaired mobility negatively impacts navigation ability and map recall in a virtual environment in persons with Multiple Sclerosis

Authors: *T. N. TAKLA[1,2], A. N. CHARGO[3,4], A. M. DAUGHERTY[3,4], N. E. FRITZ[1,5,6]; [1]Neuroimaging and Neurorehabilitation Lab., [2]Dept. of Psychiatry and Behavioral Neurosciences, [3]Dept. of Psychology, [4]Inst. of Gerontology, [5]Dept. of Hlth. Care Sci., [6]Dept. of Neurol., Wayne State Univ., Detroit, MI

Abstract: Spatial navigation is an essential component for independent living and encompasses one's ability to travel through their environment to reach a goal location. Successful real-world navigation relies on complex cognitive processing, memory of the environment and intact mobility. The role of mobility in the act of navigation is clear, however its impact on cognitive processing that supports navigation efficiency and memory of the environment is unknown. The use of virtual environments has created opportunity to explore navigation abilities in individuals with mobility impairments, including individuals with Multiple Sclerosis (MS) who experience both motor and cognitive deficits. Therefore, the objective of this study was to examine relations between mobility and virtual navigation abilities in persons with MS. We examined performance in a virtual Morris water maze while seated at a computer with a joystick and subsequent free recall of the environment details in a clinical sample of 23 ambulatory individuals with MS (18 females and 5 males, age 25 - 67 years). Mobility was assessed by clinical walking tests in the forward and backward directions. Individuals with worse mobility measured by slower backward walking velocity required longer time to navigate to the goal location in the virtual environment (r = -.493, p = .017), which suggests a mobility-related cognitive impairment in navigation ability. Though not significant, individuals with slower forward walking velocity had worse map recall of the virtual environment (r = .397, p = .067). Individuals with worse map recall had worse navigation efficiency (time: r = -.440, p = .04; distance: r = -.428, p = .047). These results highlight the unique intersection of cognition and mobility during navigation in a clinical sample of persons with MS. Given that the virtual navigation task is performed while seated, evidence of

any correlation with mobility suggests differences in cognitive processing that cannot be directly attributed to walking impairments. Future studies should examine navigation abilities and walking performance in a larger sample size and evaluate how impairments in mobility may influence cognitive processes that support navigation to better understand this complex relation.

Disclosures: T.N. Takla: None. **A.N. Chargo:** None. **A.M. Daugherty:** None. **N.E. Fritz:** None.

Poster

PSTR175. Human Navigation: Neurophysiology, Cognitive Mechanisms, Deficits

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR175.09/TT4

Topic: H.09. Spatial Navigation

Support: NRF Grant 2021M3E5D2A01023891
NRF Grant 2021M3A9E4080780
Samsung Electronics Co., Ltd (A0426-20220108)

Title: Stressor-specific modulation of hippocampal networks during spatial navigation

Authors: *K. CHOI, S. LEE;
Dept. of Brain and Cognitive Sci., Seoul Natl. Univ., Seoul, Korea, Republic of

Abstract: Context-dependent stressors impact spatial learning and memory by modulating hippocampal activity. Although most of the previous stress-related studies used paradigms in which multiple stressors are combined (e.g., social and performance-related stress), it is possible that different stressors influence the hippocampus by means of specific and distinct underlying neural networks. In this study, we investigated the effect of two types of stress conditions (time stress using instructions and indicators to hurry, and social stress using a floating video of three evaluators in a “Zoom” window scrutinizing participants’ behavior) on the hippocampus in a sample of 46 participants (mean age 24.1, 28 females). Neural activity was measured using fMRI while participants performed a spatial navigation task that required them to learn locations of objects along a given route and then navigate back to them during retrieval. Our findings revealed stressor-specific correlates of both navigation behavior and subjective stress rating. Overall, subjective stress rating was correlated with overall navigation performance and navigation strategy. Higher stress ratings were associated with longer navigation times and a tendency to follow familiar routes. While these trends were observed at the group level in the social stress condition, a high degree of individual variability was observed in the time stress condition, with some participants showing enhanced performance (compared to the “no-stress” control condition), despite the high stress ratings. Activation in the posterior hippocampus was correlated with navigation performance in all three conditions during the retrieval session, and this hippocampal activation was modulated by amygdala activation. We also found stressor-specific differences: in the time stress condition, there was a significant group-level increase in

hippocampus and amygdala activation. On the other hand, in the social stress condition, a significant decrease in striatum activation and an increase in activation of the "theory of mind" network, such as the right temporoparietal junction, were observed. Interestingly, while amygdala activation reflected subjective stress ratings in the control condition, these correlations were not significantly different in both stress conditions, suggesting that self-ratings did not reliably reflect amygdala activity in stress-related contexts. These findings provide insight into our variable vulnerability to stress by providing evidence for both stressor-specific neural networks that influence our cognition and behavior through their interactions with the amygdala and hippocampus.

Disclosures: K. Choi: None. **S. Lee:** None.

Poster

PSTR175. Human Navigation: Neurophysiology, Cognitive Mechanisms, Deficits

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR175.10/TT5

Topic: H.09. Spatial Navigation

Support: NRF Grant 2021M3E5D2A01023891
NFR Grant 2020K1A3A1A19088932
Creative-Pioneering Researchers Program through Seoul National University(SNU)

Title: Age-related Decline in Spatial Scene Order Memory in Middle-Aged Individuals

Authors: *M. HWANG[1], S.-E. PARK[1], M. CHOI[2], S. LEE[1];
[1]Seoul Natl. Univ., Seoul, Korea, Republic of; [2]Univ. of California, Berkeley, Berkeley, CA

Abstract: Hippocampal binding of objects and places across time plays an essential role in both spatial navigation and episodic memory. Previous research showed that the representation of the temporal structure of memory is more vulnerable to aging than other components (e.g., what and where) and more difficult to improve through cognitive training. In this study, we aimed to understand the age-related decline in remembering a temporal sequence of scenes and to identify its neural correlates in the hippocampus and parahippocampal regions. We performed an event-related fMRI experiment in 62 subjects (young group: n=29 (15 females, mean age 24.4 years), middle-aged group: n=33 (22 females, mean age 55.4 years). In each run, subjects were instructed to memorize 16 spatial scenes presented sequentially. After the encoding session, they were asked forced-choice questions about the objects in the scene, their spatial locations, and the order in which they saw the scenes. To exclude low-level visual signals during the recognition phase, Fourier-transformed images of the scenes used in the encoding session were added as control stimuli. In the behavioral results, temporal order memory performance showed the highest level of age-related decline. Consistent with previous studies, we found that younger subjects exhibited a significant modulation of when accuracy by the temporal distance (TD)

between the scene test pairs, there was a weak correlation between TD and behavioral accuracy in the middle-aged bad-performing group. Based on the individual variability of TD-modulated behavior in the middle-aged group, we hypothesized that the hippocampal representation of TD would be weaker in participants with bad scene order memory. First, we found that the good-performing participants showed a significant correlation between hippocampal activation and overall accuracy while the bad group did not. In the low TD trials, the good group also showed a correlation between scene order memory performance and TD-modulated activation in the entorhinal cortex. In contrast, in the high TD trials, the TD-modulated activation of the parahippocampal cortex was correlated with accuracy. The bad group did not show such TD-modulated neural activity, consistent with their behavioral patterns. These results suggest that individual differences in age-related cognitive decline related to spatial navigation and episodic memory can be partly attributed to the degeneration of hippocampal representation of the temporal sequence.

Disclosures: **M. Hwang:** None. **S. Park:** None. **M. Choi:** None. **S. Lee:** None.

Poster

PSTR175. Human Navigation: Neurophysiology, Cognitive Mechanisms, Deficits

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR175.11/TT6

Topic: H.09. Spatial Navigation

Support: NRF 2021M3E5D2A01023891
NRF 2020K1A3A1A19088932

Title: Aperiodic and periodic intracranial EEG correlates of aging and age-related spatial memory decline

Authors: *S.-E. PARK[1], T. DONOGHUE[3], J. JACOBS[3], S. LEE[2];
[1]Brain and Cognitive Sci., [2]Seoul Natl. Univ., Seoul, Korea, Republic of; [3]Columbia Univ., New York, NY

Abstract: Age-related decline in cognitive function, particularly in spatial memory, occurs even in the absence of neurodegenerative disorders. Previous neuroimaging studies have associated functional degeneration of the hippocampus with spatial memory impairment, but details of the underlying neurophysiological mechanisms have been elusive due to the co-occurrence of general age-related changes and those correlated with cognitive performance. Intracranial EEG (iEEG) recordings provide a deeper understanding of human cognition by identifying direct electrophysiological correlates of spatial memory in the hippocampus and cortical areas. Therefore, in this study, we used iEEG recordings across the whole brain to investigate age-related functional changes in neural activity in 69 presurgical epilepsy patients (19 to 61 years of age, median age 34), while they performed a computer-based spatial navigation task. Specifically, we isolated periodic narrowband power from the aperiodic broadband component

(1/f spectral slope) by applying the FOOOF (fitting oscillations & one-over f) algorithm. In the aperiodic component, we found across aging a flatter 1/f slope in the prefrontal cortex indicating an excitatory state and, in contrast, a steeper 1/f slope in the hippocampus indicating an inhibitory state. Periodic narrowband oscillations showed a frequency-dependent pattern in aging: First, we found decreases in theta (3-6Hz) power in the hippocampus. Second, while older subjects (44 to 61 years of age) showed increased alpha (6-10Hz) and beta (12-18Hz) power in various brain regions such as the hippocampus, frontoparietal and temporal cortex, gamma (37-50Hz) power showed decreases in the same brain regions. The prefrontal cortex did not show noticeable age-related changes in periodic narrowband power. When examining periodic and aperiodic components relate to behavioral performance, we first found that the decrease in hippocampal theta was more pronounced in low-performing older subjects. On the contrary, high-performing older subjects had a flatter aperiodic 1/f slope in the dorsolateral prefrontal cortex (DLPFC). The hippocampal 1/f slope was not correlated with the performance. By leveraging brain-wide spectral features using task-based iEEG, these results show both an age-related degeneration of hippocampal memory function, as indicated by the weakened theta oscillation in low-performing subjects, alongside a potential compensatory mechanism in the older subjects via flattened broadband power spectrum in DLPFC.

Disclosures: S. Park: None. **T. Donoghue:** None. **J. Jacobs:** None. **S. Lee:** None.

Poster

PSTR175. Human Navigation: Neurophysiology, Cognitive Mechanisms, Deficits

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR175.12/TT7

Topic: H.09. Spatial Navigation

Support: NRF 2021M3E5D2A01023891
Creative-Pioneering Researchers Program through Seoul National University(SNU)
Samsung Electronics Co., Ltd

Title: Discretization of space into prototype cognitive maps in human navigation

Authors: *J. KIM[1], J. SHIN[1,2], S. LEE[1];
[1]Seoul Natl. Univ., Seoul, Korea, Republic of; [2]Korea Advanced Inst. of Sci. and Technol., Daejeon, Korea, Republic of

Abstract: We integrate various types of spatial information to remember locations and navigate successfully. One powerful source of input to our cognitive maps is the environmental geometry and the spatial boundaries that comprise it. Although in real life, we navigate through complex environments with multiple goals, most studies thus far have used single target tasks. Here we tested cognitive mapping in multi-target navigation and examined how environmental geometry is used under such task demands. 17 participants (6 females, mean age 24.4) were instructed on

each trial to collect eight sequentially-presented coins while navigating in square-shaped virtual rooms. After the interference task, they were asked to retrace their path and indicate each coin's location along the way. Each participant performed a total of 65 trials across seven sessions over 2 weeks; after the final session, neural activity was recorded while they performed the task inside the fMRI scanner. First, we found that participants generally remembered locations near boundaries better but, interestingly, they also performed well at the center of the arena. Upon closer look, we found consistent response biases to nine nodes, 3 on each side of the square environment. This biased spatial pattern was similar across individuals, persisted even when they were disoriented (i.e., made large distance errors), and was invariant across different contexts and sessions. Additionally, the effect of the prototype on location memory was significant after the first coin, and increased the accuracy of the following coin, suggesting that the prototype-map boosted serial location memory by possibly increasing cognitive capacity for encoding multiple targets. fMRI scans showed a prototype-specific activation in the frontopolar cortex and the hippocampus. Moreover, at the retrieval session, the functional connectivity between the frontal lobe and the hippocampus, as well as connectivity within the frontal lobe, increased in those individuals who showed greater prototype-based biases in their response. For the first time, this study demonstrates that humans readily form and use geometry-based prototype-like cognitive maps that "chunk" space into discrete units and help them navigate. This prototype-based behavior is associated with fronto-hippocampal activation, particularly the frontopolar cortex. Further studies are needed to identify the factors that determine the characteristics of prototype maps, such as the number of nodes, scale, and individual variability in their use.

Disclosures: **J. Kim:** None. **J. Shin:** None. **S. Lee:** None.

Poster

PSTR175. Human Navigation: Neurophysiology, Cognitive Mechanisms, Deficits

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR175.13/TT8

Topic: H.09. Spatial Navigation

Support: NSF Grant 2124252

Title: Neural correlates of attention and memory for landmarks during real-world navigation

Authors: ***L. AUGUSTIN**[1], L. MACKINNEY[1], L. GARCIA[1], U. TOPALOVIC[2], M. VALLEJO MARTELO[2], M. STANGL[2], T. DAVIS[1], M. HOLLEARN[1], J. CAMPBELL[1], D. ELIASHIV[2], N. HASULAK[3], S. HILLER[2], I. FRIED[2], N. SUTHANA[2], C. INMAN[1];
[1]Univ. of Utah, Salt Lake City, UT; [2]UCLA, David Geffen Sch. of Med., Los Angeles, CA; [3]Phoenix Res. Consulting LLC, Gilbert, AZ

Abstract: Tools to understand how visual attention is allocated during real-world navigation have only recently become available. Thus, our current understanding of visual attention and memory is primarily informed by laboratory studies with stimuli on a computer screen. Because

cognitive resources are finite, visual attention to environmental stimuli like landmarks during spatial navigation must be prioritized when building a map of our prior experiences. The present study allowed us to examine how visual attention influences our ability to subsequently remember landmarks during real-world navigation and the relationship between visual attention, memory, and changes in temporal lobe (TL) oscillatory activity. Five participants implanted with NeuroPace Responsive Neurostimulators recording local field potentials (LFPs) from their TL were tasked with learning how to navigate a complex 0.75-mile route through buildings and the outdoors well enough that the participant could navigate the route in the opposite direction. Subjects walked the route 7-8 times across two days, with the 1st walk guided (encoding) and 6-7 of the walks navigated by the participant themselves (retrieval), followed by a landmark recognition task. Mobile eye-tracking and LFP data was continuously collected throughout each participant's walk synchronized with a suite of 1st person experience sensors at millisecond precision. Using this unique data, we found that allocation of visual attention and TL activity was greater for subsequently remembered versus forgotten landmarks. Taken together, these findings support our hypotheses that the amount of visual attention to landmarks during real-world navigation influences subsequent memory and modulates medial and lateral temporal lobe activity.

Disclosures: **L. Augustin:** None. **L. MacKinney:** None. **L. Garcia:** None. **U. Topalovic:** None. **M. Vallejo Martelo:** None. **M. Stangl:** None. **T. Davis:** None. **M. Hollearn:** None. **J. Campbell:** None. **D. Eliashiv:** None. **N. Hasulak:** A. Employment/Salary (full or part-time); Neuropace, Inc. (Employee). E. Ownership Interest (stock, stock options, royalty, receipt of intellectual property rights/patent holder, excluding diversified mutual funds); Stock and Stock options in NeuroPace, Inc.. **S. Hiller:** None. **I. Fried:** None. **N. Suthana:** None. **C. Inman:** None.

Poster

PSTR175. Human Navigation: Neurophysiology, Cognitive Mechanisms, Deficits

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR175.14/TT9

Topic: H.09. Spatial Navigation

Support: NIH Grant U01NS103802
NIH Grant U01NS103780
NIH Grant K99NS126715
McKnight Foundation
Keck Foundation

Title: Differential saccadic modulation of theta and gamma oscillatory activity in the human medial temporal lobe during real-world spatial navigation with and without memory

Authors: ***H. N. ZUBAIR**[1], M. STANGL[2], U. TOPALOVIC[2], C. S. INMAN[5], S. HILLER[2], V. R. RAO[6], C. H. HALPERN[7], D. ELIASHIV[3], I. FRIED[8], N. A. SUTHANA[4];

[1]David Geffen Sch. of Med., UCLA Chapter, Los Angeles, CA; [3]Dept. of Neurol., [4]Neurosurg. / Psychiatry / Psychology, [2]UCLA, Los Angeles, CA; [5]Psychology, Univ. of Utah, Salt Lake City, UT; [6]Neurol., Univ. of California, San Francisco, San Francisco, CA; [7]Neurosurg., Hosp. of the Univ. of Pennsylvania, Philadelphia, PA; [8]Neurosurg., UCLA Sch. Med., Los Angeles, CA

Abstract: Humans use saccadic eye movements to explore and learn important locations within their environment. How saccades influence neural activity within the human medial temporal lobe (MTL) during real-world spatial navigation has not been investigated, largely due to technical challenges associated with recording deep brain activity during physical movement. In this study, intracranial electroencephalographic (iEEG) activity was recorded simultaneously with body and eye movements in humans during freely-moving exploration of a real-world environment. Participants alternated between two tasks: 1) Visually-cued navigation, which consisted of walking towards one of 20 unique visible wall-mounted signs and 2) Memory-cued navigation, which consisted of walking towards and remembering hidden target locations within the room. MTL low frequency (3-14 Hz) oscillatory activity significantly increased during saccades compared to fixations while navigating the environment. Interestingly, increases in MTL theta (6-8 Hz) bandpower during saccades were specific to successful memory-cued navigation and not to low performance or visually-cued navigation. Saccade magnitude was quantified as the two-dimensional displacement of the eye between start and end of the saccade. In memory-cued navigation, theta power steadily increased during the saccade and was overall elevated in saccades of high- vs. low- magnitude. Conversely, visually-cued navigation featured an increase in gamma (45-80 Hz) oscillatory activity, particularly in the 40 ms surrounding saccade onset. Altogether, these findings show that saccadic eye movements modulate MTL theta and gamma power during real-world ambulatory spatial navigation.

Disclosures: H.N. Zubair: None. **M. Stangl:** None. **U. Topalovic:** None. **C.S. Inman:** None. **S. Hiller:** None. **V.R. Rao:** None. **C.H. Halpern:** None. **D. Eliashiv:** None. **I. Fried:** None. **N.A. Suthana:** None.

Poster

PSTR175. Human Navigation: Neurophysiology, Cognitive Mechanisms, Deficits

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR175.15/TT10

Topic: H.09. Spatial Navigation

Support: U01NS117838 , NIH NINDS

Title: Theta oscillations are temporally structured during real-world and imagined navigation in humans

Authors: *M. SEEBER[1], M. STANGL[1], M. VALLEJO[1], U. TOPALOVIC[1], S. HILLER[1], C. H. HALPERN[2], J.-P. LANGEVIN[1], V. R. RAO[3], I. FRIED[1], D. ELIASHIV[1], N. SUTHANA[1];

[1]UCLA, Los Angeles, CA; [2]Univ. of Pennsylvania, Philadelphia, PA; [3]Univ. of California, San Francisco, San Francisco, CA

Abstract: Neural mechanisms of spatial navigation are considered evolutionary precursors of episodic memory. Given that recent neurotechnological developments have enabled the study of real-world navigation in humans, we were able to investigate neural oscillations recorded from a chronically implanted device in five individuals receiving responsive neurostimulation therapy. Participants were asked to walk two different trajectories, including four turns each, in an indoor room (14.6×13.5 m^2). Feedback on performance was provided on a tablet screen to refine their walking trajectories after each run by overlaying their actual movement trajectories recorded with motion capture on the ideal, instructed routes. After each actual walk, participants walked on a treadmill while imagining these walking trajectories in their minds. During this real-world spatial navigation task, intracranial electroencephalography (iEEG) was recorded from electrodes located in the medial temporal lobe (MTL). Theta oscillations were evident in each participant at spectral peaks in the 3-10 Hz frequency range, in agreement with previous reports. Herein, we studied the temporal structure of theta envelopes since theta oscillations have been shown to occur in short-lasting bouts in humans. After aligning motion trajectories to each other, we accordingly investigated theta power fluctuations along the participants' trajectories. Single trials showed theta bouts frequently occurring at upcoming turns. This temporal alignment resulted in theta dynamics resembling the maze structure and was consistent across trials (30-35, left/right walks each). Furthermore, these theta dynamics were apparent in each participant and were significantly higher than chance as assessed by permutation data with abolished time relation between iEEG and motion capture data. Similarly, we found theta bouts occurring at specific time points during imagined navigation but not during sole treadmill walking. These temporally structured theta bouts were present at the same anatomical locations during actual and imagined navigation, suggesting that similar functional networks are involved in both types of navigation. Visual flow was absent during imagined navigation and bodily cues were identical to sole treadmill walking. Therefore, these findings demonstrate the capability of the MTL to internally generate theta dynamics relevant for memory retrieval in the absence of external sensory cues. Altogether, this study might contribute to a better understanding of neural mechanisms providing temporal structure to organize complex experiences into well-defined segments or mnemonic episodes.

Disclosures: **M. Seeber:** None. **M. Stangl:** None. **M. Vallejo:** None. **U. Topalovic:** None. **S. Hiller:** None. **C.H. Halpern:** None. **J. Langevin:** None. **V.R. Rao:** None. **I. Fried:** None. **D. Eliashiv:** None. **N. Suthana:** None.

Poster

PSTR175. Human Navigation: Neurophysiology, Cognitive Mechanisms, Deficits

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR175.16/TT11

Topic: H.09. Spatial Navigation

Support: National Science Foundation IIS-2024633

Title: Individual differences in navigation: survey and graph knowledge

Authors: *N. KAUSHIK[1], A. TU[1], M. HEGARTY[2], E. R. CHRASTIL[1];
[1]Univ. of California, Irvine, Irvine, CA; [2]Univ. of California, Santa Barbara, Santa Barbara, CA

Abstract: Two of the most common spatial navigation strategies are graph and survey knowledge. Graph knowledge refers to the topographical understanding of how different locations are connected, and survey knowledge refers to the understanding of the distances and angles between locations. In current literature, it has been noticed that differences in individual ability in wayfinding strategies, due to the individualized need for executing paths and reaching goal locations. However, it is unclear the relationship between individual differences in survey and graph knowledge, and how this relationship is affected by other factors like time or repeated practice. Based on past research, we hypothesize that there will be a positive correlation between survey and graph knowledge, indicating that high accuracy in one type of knowledge will indicate high accuracy in the other. To test this hypothesis, we test participants' spatial knowledge using virtual reality mazes and testing. Participants are given 3 minutes to explore the maze to learn the location of 8 objects. After, they are tested in two different ways, as means of testing both types of spatial knowledge. The pointing task tests survey knowledge, where participants are instructed to point in the angle of another object while facing one of the objects. The wayfinding task tests graph knowledge where participants start at one object and are instructed to walk to another object. Our analysis found a significant correlation between survey and graph accuracy, with repeated practice with the test trials having no significant effect. We also observed that more time spent per trial resulted in less accurate wayfinding on average. We further analyzed sex differences for the correlation between survey and graph knowledge, finding that both male and female correlations are similar to the entire data set. Given this preliminary analysis, we intend to further represent the data in different models to better understand how sex differences play a role in the individual differences in navigation. Together, these findings provide insight into the relationship between different types of spatial knowledge and into individual differences in navigation abilities.

Disclosures: N. Kaushik: None. **A. Tu:** None. **M. Hegarty:** None. **E.R. Chrastil:** None.

Poster

PSTR175. Human Navigation: Neurophysiology, Cognitive Mechanisms, Deficits

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR175.17/TT12

Topic: H.09. Spatial Navigation

Support: NIH Grant T32AG073088
NIH Grant F32NS129626
UCSB Faculty Senate Grant

Title: Brain networks supporting egocentric and allocentric relational processing

Authors: *M. J. STARRETT AMBROSE[1], Y. CHENG[2], R. C. DAVIS[4], B. TRANQUADA-TORRES[1], E. R. CHRASTIL[1,3];
[1]Neurobio. and Behavior, [2]Cognitive Sci., [3]Ctr. for the Neurobio. of Learning and Memory, Univ. of California, Irvine, Irvine, CA; [4]Geography, Univ. of California, Santa Barbara, Santa Barbara, CA

Abstract: The ability to orient oneself within an environment is essential for successful navigation. However, the utility of a cognitive capacity for "orienting" extends beyond the realm of spatial processing. Indeed, humans use figural representations to illustrate spatial (maps), temporal (timelines), and even social (family trees or personal networks) relationships, whereby a one-dimensional "distance" can be ascribed to the relationships between any two "points." Recent research investigating the neural mechanisms supporting non-spatial relational processing suggest that there is substantial overlap with canonical spatial processing networks (i.e., domain general function), although important distinctions may still exist (domain specific function). To-date, few studies have provided a systematic investigation into the degree of overlap between conceptually separable cognitive domains and their corresponding brain networks. Moreover, no study has explicitly incorporated manipulations relating to a core feature of canonical spatial processing networks: the distinction between self-referential (egocentric) and observer-independent (allocentric) frames of reference. To address this, we developed a task that directly compares distance ratings across domains (spatial, temporal, and social) and reference frames (egocentric and allocentric) frames. Participants were cued whether a judgement would be spatial, temporal, or social, then viewed side-by-side images of one well-known person and, crucially, either the participant themself (egocentric) or a different well-known person (allocentric) followed by a one-to-seven distance rating for the cued domain. Critically, the same pairs were used for all domains, thus equating the stimuli while only varying the type of judgment. Participants completed eight blocks of thirty trials during fMRI scanning and then rated their confidence for each judgment outside the scanner. In ongoing work, we are using functional magnetic resonance imaging (fMRI) to infer a neural index of subjective distances across domains and reference frames. By including both egocentric and allocentric judgements, we are uniquely positioned to compare brain networks that facilitate temporal and social relational processing with long-standing spatial models of how the brain individually represents and translates between frames of reference.

Disclosures: M.J. Starrett Ambrose: None. **Y. Cheng:** None. **R.C. Davis:** None. **B. Tranquada-Torres:** None. **E.R. Chrastil:** None.

Poster

PSTR175. Human Navigation: Neurophysiology, Cognitive Mechanisms, Deficits

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR175.18/TT13

Topic: H.09. Spatial Navigation

Support: NIH T32-GM136624

Title: Brain network connectivity and dynamics of navigational learning and memory

Authors: *E. WARD[1], R. WOODRY[2], J. M. CARLSON[3], E. R. CHRASTIL[1];
[1]Univ. of California, Irvine, Irvine, CA; [2]New York Univ., New York, NY; [3]Dept. of Physics, Univ. of California, Santa Barbara, Santa Barbara, CA

Abstract: Brain Network Connectivity and Dynamics of Navigational Learning and Memory
Erica Ward, Rob Woodry, Jean Carlson, Elizabeth Chrastil
Effective spatial navigation is dependent on several cognitive processes including learning, attention, and memory. But how do people learn and remember new environments? Several studies have explored the brain activity of already known environments, however, comparatively little is known about the acquisition of this knowledge. In the present study, healthy young adults (n=82) completed a challenging maze learning task during fMRI scanning. Participants were given 16 minutes to explore a virtual hedge maze and learn the locations of 9 objects. Next, their object location memory was tested in a series of 48 (45 sec) trials, each of which started at one object with instructions to find another object using the paths of the maze (e.g. clock lamp). To reduce feedback in the testing phase, all objects were replaced with red spheres. Accuracy ranged from near 0% to 100% correct, enabling us to quantify brain network and behavioral differences that distinguish between poor, average, and exceptional performers. We used dynamic community detection to identify brain network changes during the learning and test phases, and to determine whether the network dynamics differentiate between good and poor learners. We parcellated the brain images into ROIs (Schaefer 200 atlas combined with the Harvard-Oxford subcortical atlas) and calculated coherence values between each ROI. Then, by assigning each ROI to an algorithm-determined community in each time window, we determined whether and how each ROI changed networks over time. Specifically, we examined flexibility–the number of times an ROI changed communities. Using these metrics, we characterized the dynamic brain networks during the learning and test phases. Preliminary results suggest that the best navigators exhibited high flexibility throughout the brain as related to performance, whereas average and poor navigators exhibited low flexibility only in the salience network. To complement this novel approach, we conducted a traditional functional connectivity analysis to further quantify connectivity between key brain regions, such as the hippocampus, prefrontal cortex, and striatum, throughout the learning and test phases. Together, this study provides greater understanding of changes in brain communication when navigating in a new environment, lending insight into the dynamics of learning more broadly.

Disclosures: E. Ward: None. **R. Woodry:** None. **J.M. Carlson:** A. Employment/Salary (full or part-time); University of California, Santa Barbara. **E.R. Chrastil:** A. Employment/Salary (full or part-time); University of California, Irvine.

Poster

PSTR175. Human Navigation: Neurophysiology, Cognitive Mechanisms, Deficits

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR175.19

Topic: H.09. Spatial Navigation

Support: Merck Investigator Studies Program grant MISP-57175
Alzheimer's Society grants 178, 264 and 397
UK National Institute for Health Research Clinical Research Network and Biomedical Research Centre Cambridge grant 1215-20014
US Alzheimer's Association grant TriBEKa-17–519007
Wellcome grant 098436/Z/12/B
Wellcome grant 202805/Z/16/Z
UK Medical Research Council grant SUAG/046 G101400
National Institute of Neurological Disorders and Stroke of the National Institutes of Health grant K99NS126715

Title: Impaired path integration may herald disease onset in Alzheimer's disease

Authors: *C. NEWTON[1,2], M. POPE[2,3], C. RUA[2], R. N. HENSON[4], Z. JI[1], N. BURGESS[1], C. RODGERS[2], M. STANGL[5], M. DOUNAVI[2], A. CASTEGNARO[1], I. KOYCHEV[6], P. MALHOTRA[7], T. WOLBERS[8], K. RITCHIE[9], C. RITCHIE[10], J. O'BRIEN[2,3], L. SU[2], D. CHAN[1];
[1]Univ. Col. London, London, United Kingdom; [2]Univ. of Cambridge, Cambridge, United Kingdom; [3]Cambridgeshire Peterborough NHS Fndn. Trust, Cambridge, United Kingdom; [4]MRC CBU, Univ. of Cambridge, Cambridge, United Kingdom; [5]UCLA, Los Angeles, CA; [6]Univ. of Oxford, Oxford, United Kingdom; [7]Imperial Col. London, London, United Kingdom; [8]DZNE, Magdeburg, Germany; [9]Inserm, Inst. de Neurosciences, Montpellier, France; [10]Univ. of Edinburgh, Edinburgh, United Kingdom

Abstract: Introduction. Detecting the onset of cognitive impairment in Alzheimer's disease (AD) is key to maximising the success of future disease-modifying drugs. The entorhinal cortex (EC) is the first cortical site of AD neurodegeneration, with pathological spread associated with EC grid cell dysfunction. Given the role of grid cells in path integration-based navigation (PI) we predicted that impaired path integration would represent the first behavioural change in adults at risk of AD. **Method.** One hundred asymptomatic midlife adults with either hereditary (family history or APOE-ε4 allele) or physiological AD risk factors (the Cardiovascular Risk Factors, Aging and Dementia Study (CAIDE) risk score) undertook an immersive virtual reality PI task alongside testing of other domains previously reported sensitive to preclinical AD. These included probes of episodic memory, allocentric and egocentric spatial processing, which we compared to PI using receiver operator characteristics analysis. In n=55, ultra-high field 7T multimodal MRI was used to assess i) the volume of brain regions of interest associated with both navigation and early AD and ii) an fMRI BOLD response modulation reflecting grid-like signals in the EC. **Results.** Impaired PI selectively predicted both hereditary and physiological AD risk, with no corresponding effects on the comparator cognitive tests. Post-hoc pairwise tests showed family history and APOE-e4 risk PI impairments were specific to males, whereas females with a family history showed poorer egocentric processing. PI impairments across all risk factors were related to poorer angular estimation in the absence of visual orientation cues, and in males, associated with negative hexadirectional grid-like fMRI signals in the posterior-medial EC. Further analysis of these negative signals revealed evidence of a unimodal head-

direction-like fMRI signal, which we consider as a compensatory mechanism for functional changes to hexadirectional grid-cell signals due to incipient AD pathology. **Conclusion.** Disruption of path integration in people at risk of AD may predate impairment in other cognitive domains, notably episodic memory, and as such may represent the all-important transition point from at-risk status to clinical disease onset. This is critical for early disease detection. As well as benefiting clinical practice, the application of a PI test based on grid cell function aids translational research in providing a platform by which AD changes at the cellular level may be linked to the clinical onset of the disease.

Disclosures: **C. Newton:** None. **M. Pope:** None. **C. Rua:** None. **R.N. Henson:** None. **Z. Ji:** None. **N. Burgess:** None. **C. Rodgers:** None. **M. Stangl:** None. **M. Dounavi:** None. **A. Castegnaro:** None. **I. Koychev:** None. **P. Malhotra:** None. **T. Wolbers:** None. **K. Ritchie:** None. **C. Ritchie:** None. **J. O'Brien:** None. **L. Su:** None. **D. Chan:** None.

Poster

PSTR176. Animal Navigation

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR176.01/TT14

Topic: H.09. Spatial Navigation

Title: Egocentric spatial representations of boundaries and rewards in dorsomedial striatum

Authors: M. RASHED-AL-MAHFUZ[1], R. R. ROZESKE[2], J. R. HINMAN[1];
[1]Dept. of Psychology, Univ. of Illinois Urabana-Champaign, Champaign, IL; [2]Psychology, Univ. of Toronto, Toronto, ON, Canada

Abstract: The overt behavior of navigation requires representation of the environment in an egocentric reference frame, as this is the way all organisms move through the world. Sensory information is represented in an egocentric reference frame, which is utilized to generate the allocentric spatial representation of the hippocampal formation. The allocentric representation must then be transformed back into an egocentric reference frame to direct ongoing behavior. The dorsomedial striatum (DMS) serves as a critical node in directing the ongoing behavior of animals and has previously been shown to represent spatial information in an egocentric reference frame.
We imaged hundreds of single cells simultaneously (34 - 294 cells/mouse) using 1-photon calcium imaging to understand the spatial and self-motion representations of neurons in DMS of mice (n=6) as they freely foraged in a 60 cm^2 open field. A generalized linear model (GLM) with allocentric, self-motion, and egocentric predictors allowed the identification of cells with various coding properties including egocentric boundary cells (EBCs; 8%) that respond when mice were located at a specific distance and orientation to the environmental boundaries. Cells with stable allocentric (4%) and self-motion (9%) coding were present in the DMS, with additional cells possessing conjunctive coding of some combination of those three variables. In some cases, EBCs demonstrated differences in response properties between two halves of a recording,

resulting in identification as a significant EBC during one half but not the other half. We trained the GLM with predictors from the significant EBC-half and predicted spike trains using predictors of the non-EBC half which highlights that inadequate egocentric behavioral sampling across recording halves resulted in the inability to detect the EBC given the observed behavior. This highlights an important experimental constraint when investigating egocentric spatial coding, that complete allocentric coverage of an environment does not entail sufficient egocentric sampling as well. Given sufficient egocentric sampling, EBCs show high stability both within, as well as across different environments. Finally, the responses of DMS neurons were investigated while mice foraged for a reward at a fixed site within the open field in order to identify egocentric goal vector coding. A similar GLM approach was employed again resulting in the identification of a combination of coding strategies by DMS neurons. Overall, the DMS contains a variety of navigationally relevant cells with questions outstanding about the downstream basal ganglia recipients of said information.

Disclosures: M. Rashed-Al-Mahfuz: None. **R.R. Rozeske:** None. **J.R. Hinman:** None.

Poster

PSTR176. Animal Navigation

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR176.02/TT16

Topic: H.09. Spatial Navigation

Support: NIH Grant 1R01NS11128
BRAIN Initiative Grant RF1NS113287
BRAIN Initiative Grant RF1NS126044
Minnesota Robotics Institute (MnRI)
MnDRIVE RSAM
The McKnight Foundation
UMN Mechanical Engineering Department

Title: Brain-wide neural recordings in mice navigating physical spaces enabled by a cranial exoskeleton

Authors: J. HOPE[1], *T. M. BECKERLE[4], P.-H. CHENG[1], Z. VIAVATTINE[1], M. FELDKAMP[1], S. FAUSNER[1], K. SAXENA[1], E. KO[1], I. HRYB[1], R. CARTER[2], T. J. EBNER[2], S. B. KODANDARAMAIAH[1,2,3];
[1]Mechanical Engin., [2]Neurosci., [3]Biomed. Engin., Univ. of Minnesota, Minneapolis, MN;
[4]Mechanical Engin., Univ. of Minnesota, Twin Cities, Saint Paul, MN

Abstract: Complex behaviors are mediated by neural computations occurring throughout the brain. In recent years, tremendous progress has been made in developing technologies that can record neural activity at cellular resolution at multiple spatial and temporal scales. However, these technologies are primarily designed for studying the mammalian brain during head fixation

- wherein the behavior of the animal is highly constrained. Miniaturized devices for studying neural activity in freely behaving animals are largely confined to recording from small brain regions owing to performance limitations. We present a cranial exoskeleton that assists mice in maneuvering neural recording headstages that are orders of magnitude larger and heavier than the mice, while they navigate physical behavioral environments. Force sensors embedded within the headstage are used to detect the mouse's milli-Newton scale cranial forces which then control the x, y, and yaw motion of the exoskeleton via an admittance controller. We discovered optimal controller tuning parameters that enable mice to locomote at physiologically realistic velocities and accelerations while maintaining natural walking gait. Mice maneuvering headstages weighing up to 1.5 kg can make turns, navigate 2D arenas, and perform a navigational decision-making task with the same performance as when freely behaving. We designed an imaging headstage and an electrophysiology headstage for the cranial exoskeleton to record brain-wide neural activity in mice navigating 2D arenas. The imaging headstage enabled recordings of Ca2+ activity of 1000s of neurons distributed across the dorsal cortex. The electrophysiology headstage supported independent control of up to 4 silicon probes, enabling simultaneous recordings from 100s of neurons across multiple brain regions and multiple days during a navigational decision-making task. Advances in sensor technology, data acquisition, and control algorithms are being explored to improve the interaction between the subject and the robot with a goal of more natural movement and behavior. Cranial exoskeletons provide flexible platforms for largescale neural recording during the exploration of physical spaces, a critical new paradigm for unraveling the brain-wide neural mechanisms that control complex behavior.

Disclosures: **J. Hope:** None. **T.M. Beckerle:** None. **P. Cheng:** None. **Z. Viavattine:** None. **M. Feldkamp:** None. **S. Fausner:** None. **K. Saxena:** None. **E. Ko:** None. **I. Hryb:** None. **R. Carter:** None. **T.J. Ebner:** None. **S.B. Kodandaramaiah:** E. Ownership Interest (stock, stock options, royalty, receipt of intellectual property rights/patent holder, excluding diversified mutual funds); Co-founder of Objective Biotechnology Inc.

Poster

PSTR176. Animal Navigation

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR176.03/TT17

Topic: H.09. Spatial Navigation

Support: NIH CRCNS Grant Project Number: 1R01AG076198-01

Title: A reverse translational approach enabling the assessment of navigational deficits in early Alzheimer's disease

Authors: *S. DUNCAN, D. LAYFIELD, I. CHOI, S. RAMLO, S. REHMAN, K. STICKEL, E. L. NEWMAN;
Sch. of Psychological and Brain Sci., Indiana Univ., BLOOMINGTON, IN

Abstract: Alzheimer's disease (AD) is a growing problem in the aging population. While effective treatments are still being sought, it is widely agreed that any future treatment would be more effective if administered early in disease progression. Spatial disorientation is one of the earliest clinically observable signs of AD. Therefore, the development of sensitive and specific tests of spatial memory may be a plausible method for the early detection of AD-related deficits Path integration is a cognitive mechanism underlying spatial memory involving navigation using self-motion cues (i.e. number of steps taken) in the absence of environmental landmarks. In humans, the triangle completion task is the most commonly used test of path integration, wherein subjects are asked to walk from location A to location B followed by location C before being asked to navigate back to location A, in the absence of environmental landmarks. Currently, however, we lack an analogous task used to test path integration in rodents. At present, rodent path integration is assessed using various behavioural tasks, both in open-field environments and virtual reality environments. In the present study, we developed a novel rodent version of the triangle completion task in a circular open field arena with wall-mounted liquid reward dispensers taking the place of locations A, B and C, as described above in the human version of the task. Rats were guided to each reward dispenser in sequence, by a dispenser-mounted LED, before returning the location A without any visual cues (rats were trained to return to the first dispenser in response to an audio tone). We utilized a transgenic rat model of AD (F344Tg-AD; Cohen, 2013, n = 11) to assess AD-associated deficits in this version of the triangle completion task through performance comparisons with wild-type littermates (n = 7). It is, of course, essential that the relationship between deficits observed in rodent models of AD are comparable to those observed in human AD patients. Therefore, we examined AD associated path integration deficits in a rodent analogue of the triangle completion task, allowing for more valid comparisons to path integration performance in AD, and early stage AD, human patients

Disclosures: S. Duncan: None. **D. Layfield:** None. **I. Choi:** None. **S. Ramlo:** None. **S. Rehman:** None. **K. Stickel:** None. **E.L. Newman:** None.

Poster

PSTR176. Animal Navigation

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR176.04/TT18

Topic: H.09. Spatial Navigation

Support: NIH NINDS R01 NS121413
AFOSR FA9550-14-1-0398
ONR N00014-17-1-2736

Title: Hippocampal representation of target distance in the echolocating big brown bat, Eptesicus fuscus

Authors: X. YIN[1], A. KRISHNA[1], H. LEE[2], C. YU[1], *C. F. MOSS[3];
[1]Psychological and Brain Sci., [2]Neurosci., [3]Psychological and Brain Science, Neuroscience, Kavli Neurosci. Discovery Inst., Johns Hopkins Univ., Baltimore, MD

Abstract: The hippocampal formation has been implicated in a wide range of functions, from spatial memory to time measurement. Past research has emphasized the separate functions of place cells and time cells, but these dimensions may also operate jointly in animals that rely on active sensing for target ranging. For example, the echolocating bat emits high frequency sounds and processes the time delay between calls and echoes to estimate object distance. It uses this spatiotemporal information to intercept flying insect prey and to steer around obstacles. The echolocating bat therefore presents a powerful model system to investigate hippocampal representation of space and time. Here, we present new hippocampal data from the echolocating bat, *Eptesicus fuscus*, as it performed a sonar target tracking task in the dark. Bats were trained to perch on a platform and track a target (cluster of tethered mealworms) that moved towards and away from the bat at a starting distance of 3m. The location of the platform was changed over trials to evaluate spatial coding in allocentric and egocentric reference frames. The bat's echolocation calls were recorded with an array of microphones, and its head direction was measured using a four-camera, high-speed motion tracking system. Multichannel neural recordings were taken from hippocampal CA1 of 4 behaving bats and synchronized with audio and video data. Spikes were sorted offline, and response areas of ~120 single object distance-tuned neurons were quantified. Our data show that a population of hippocampal CA1 neurons encodes the call-echo time delay/distance to an object. A subset of neurons encoding target distance responded selectively to the direction of target movement. During trial segments when the bat ceased echolocating, neural firing rate decreased dramatically, suggesting that hippocampal activity is modulated by the bat's call production and echo reception. These findings offer new insights to spatiotemporal hippocampal representation in animals actively tracking a target moving along the distance axis.

Disclosures: X. Yin: None. **A. Krishna:** None. **H. Lee:** None. **C. Yu:** None. **C.F. Moss:** None.

Poster

PSTR176. Animal Navigation

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR176.05/TT19

Topic: H.09. Spatial Navigation

Support: Simons Foundation
Office of Naval Research
Faculty Scholars Program, Howard Hughes Medical Institute

Title: Rapid goal-directed learning without catastrophic forgetting in multiple Morris watermazes

Authors: *R. L. WANG[1], J. HWANG[2], A. BOOPATHY[2], I. R. FIETE[1];
[1]Brain and Cognitive Sci., [2]EECS, MIT, Cambridge, MA

Abstract: Animals can learn new tasks and transfer knowledge between similar tasks, while retaining their abilities on previously learned tasks. By contrast, machine learning models struggle to do so. We consider how animals could learn the classic Morris Water Maze spatial reinforcement learning task, and how they are able to generalize in three ways: 1) Finding a fixed goal from new starting locations in a given environment, 2) Rapidly adjusting to new goal locations in the same environment, 3) Learning new environments without forgetting old ones. We propose a novel model, which combines a hippocampal-entorhinal-neocortical neural circuit model (vector-HASH) with a spatially invariant action (policy) network, and show that it can perform rapid goal-directed learning in new environments without forgetting old ones. This neural circuit, which we call policy vector HASH (pvHASH), exhibits all three forms of generalization without forgetting shown by animals. This work represents the first type of computational modeling for this type of task and provides insights on how structured organization in the brain can result in stronger performance (higher data efficiency, better transfer, greater resistance to forgetting) on difficult problems than networks with less structure and fewer constraints.

Disclosures: R.L. Wang: None. **J. Hwang:** None. **A. Boopathy:** None. **I.R. Fiete:** None.

Poster

PSTR176. Animal Navigation

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR176.06/TT20

Topic: H.09. Spatial Navigation

Support: NIH Grant T32GM007276
NSF Grant DGE-1656518
NSF Grant DGE-1828993

Title: Applying novel techniques to study neural correlates of amphibian spatial cognition

Authors: *D. SHAYKEVICH[1], G. A. WOODS[2], D. PAREJA MEJÍA[3], G. HONG[4], L. A. O'CONNELL[5];
[2]Applied Physics, [1]Stanford Univ., Stanford, CA; [3]Biol., Stanford Univ., Stanford University, CA; [4]Materials Sci. and Engin., [5]Biol., Stanford Univ., Stanford, CA

Abstract: Many animals perform tasks requiring the processing of spatial information. Modern neuroscience tools have allowed for in depth characterization of space coding neurons (e.g. place and grid cells) in the mammalian hippocampus and entorhinal cortex, enabling a rich understanding of how organisms navigate and remember environments. Much less work has characterized cells with similar functions in other vertebrates, especially in amphibians. We are performing field- and lab-based research in the cane toad, *Rhinella marina*, a large and globally

abundant amphibian, to study how the amphibian brain supports navigation and encodes spatial information. Our main target is the medial pallium, the proposed amphibian homolog of the mammalian hippocampus, which we hypothesize is activated during navigation and contains spatial coding neurons.

First, we completed a translocation-homing study of invasive cane toads in Hawai'i and demonstrated that toads can return home from distances exceeding 1 km. We collected brains from toads and quantified cells immunoreactive for a marker of neural activation and identified pallial brain regions that are activated during homing (1A).

In addition, we developed tools that allow for recording of spatial neural coding. Typical electrophysiology experiments in amphibians have been limited by the movement of the brain, which has led to most recordings requiring paralysis. Here, we show preliminary results for the implementation of flexible mesh electronics in toads (1B). Mesh electronics have been used in rodent studies to produce stable recordings with single-unit resolution. The electronics' bioinspired properties, combined with their robust electrical connection (1C), enable our unprecedented study of awake, behaving toads. We have performed mock surgeries (1D) and developed a toad-specific surgery protocol to allow for mesh implantation. Ultimately, translating this technology in behaving toads will enable us to identify spatial encoding neurons, potentially revealing novel mechanisms for learning and navigation beyond mammalian models.

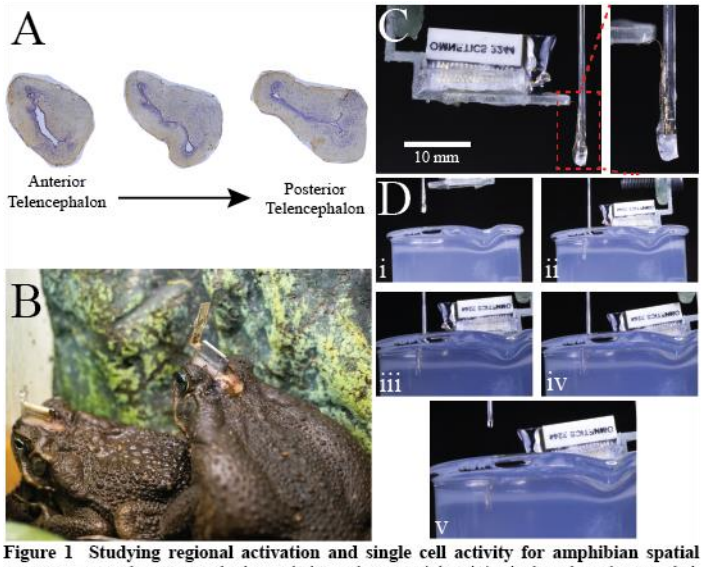


Figure 1 Studying regional activation and single cell activity for amphibian spatial cognition. (A) The cane toad telencephalon, where spatial activity is thought to be encoded. **(B)** Cane toads implanted with flexible mesh electronics. **(C)** Flexible mesh electronics loaded on a surgical insertion shuttle and bonded to omnetics connector. Inset shows mesh attached to shuttle with polyethylene glycol (PEG). **(D)** Mock insertion in hydrogel showing (i) mesh loaded on the shuttle aboive the hydrogel phantom, then (ii) lowered into the hydrogel where the PEG dissolves (iii-iv) and the shuttle is retracted (v) while the mesh remains in the hydrogel.

Disclosures: D. Shaykevich: None. **G.A. Woods:** None. **D. Pareja Mejía:** None. **G. Hong:** None. **L.A. O'Connell:** None.

Poster

PSTR176. Animal Navigation

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR176.07/TT21

Topic: H.09. Spatial Navigation

Support: NIH Grant K99EY033850
Howard Hughes Medical Institute

Title: The superior colliculus is critical for virtual head turns and eye movements during REM sleep

Authors: *Y. SENZAI[1], M. SCANZIANI[2];
[2]Univ. of California, San Francisco, [1]Univ. of California, San Francisco, San Francisco, CA

Abstract: Since the discovery of REM sleep, the nature of the rapid eye movements that characterize this sleep phase has remained elusive. Do they reveal gaze shifts in the virtual world of dreams or simply reflect random brainstem activity? In a previous study, we harnessed the head direction (HD) system of the mouse thalamus, a neuronal population whose activity reports, in awake mice, their actual HD as they explore their environment and, in sleeping mice, their virtual HD. We discovered that the direction and amplitude of rapid eye movements during REM sleep reveal the direction and amplitude of the ongoing changes in virtual HD, i.e. virtual head turns. What coordinates the direction of rapid eye movements with that of virtual head turns during REM sleep? We have tested the role of the superior colliculus (SC) because, in awake animals, the SC coordinates eye and head movements to generate gaze shifts. We have discovered that the SC activity can predict the direction of rapid eye movements and virtual head turns during REM sleep. Furthermore, we have also discovered that silencing the SC has a major impact on virtual head turns during REM sleep. These discoveries suggest that the SC, by orchestrating sensorimotor representation in the sleeping brain, may mediate gaze shifts in the virtual world of REM sleep.

Disclosures: Y. Senzai: None. **M. Scanziani:** None.

Poster

PSTR176. Animal Navigation

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR176.08/TT22

Topic: H.09. Spatial Navigation

Support: MEXT/JSPS KAKENHI 19H03538
MEXT/JSPS KAKENHI 21K15006

Title: Loss of Akain1, an endogenous micropeptide that inhibits PKA localization, attenuates contextual discrimination and behavioral flexibility in mice

Authors: K. FUJII[1,2,3], Y. KOSHIDAKA[2], M. ADACHI[2], Y. YANAGIBASHI[2], M. MATSUO[2], S. HONGO[2], H. NISHIZONO[8], I. TAKASAKI[4], N. KUROSAWA[5,6], Y. AIZAWA[9], ***K. TAKAO**[7,1,2,3,6]**;**
[1]Fac. of Med., [2]Life Sci. Res. Ctr., [3]Res. Ctr. for Idling Brain Sci., [4]Dept. of Pharmacology, Fac. of Engin., [5]Dept. of Life Sci. and Bioengineering, Fac. of Engin., [6]Ctr. for Advanced Antibody Drug Develop., [7]Univ. of Toyama, Toyama, Japan; [8]Med. Res. Inst., Kanazawa Med. Univ., Kahoku, Japan; [9]Sch. of Life Sci. and Technol., Tokyo Inst. of Technol., Yokohama, Japan

Abstract: By definition, long noncoding RNAs (lncRNAs) contain no protein-coding open reading frames (ORFs). Recent bioinformatics and high-throughput sequencing studies, however, demonstrated that many lncRNAs possess short “non-canonical” ORFs (sORFs) encoding micropeptides. These newly identified micropeptides may help to elucidate the mechanisms underlying vital phenomena and are potential targets for drug discovery. A-kinase anchor inhibitor 1 (Akain1) is a protein kinase A (PKA)-binding micropeptide encoded by an sORF. Specific PKA actions are achieved by controlling its cellular localization through a family of A-kinase anchoring proteins (AKAPs). AKAPs localize PKA to specific intracellular sites and spatially restrict intracellular signaling events. In PKA signaling, Akain1 competes with other AKAPs (e.g., AKAP1 and MAP2) to bind PKA and seems to cancel the intracellular localization of PKA. AKAP-PKA interactions control various cellular processes, including the regulation of neuroplasticity, and Akain1 mRNA is preferentially expressed in neural tissues. Whether Akain1 is translated into an endogenous protein in vivo and how Akain1 affects brain function and behavior, however, remain unclear.In this study, we generated Akain1 reporter mice and Akain1 knockout (KO) mice on the C57BL/6J background using the CRISPR-Cas9 genome editing system. Utilizing Akain1 reporter mice in which an epitope-tag coding sequence was inserted before the stop codon, we detected epitope-tag-specific cells in the mouse central nervous system (e.g., cortex, superior colliculus, cerebellum, etc.). These findings suggest that Akain1 is translated into an endogenous protein. To investigate the function of Akain1 in the brain, we subjected Akain1 deletion mice to a comprehensive behavioral test battery. In the pattern separation test, Akain1 KO mice exhibited impaired performance in distinguishing between two similar contexts. In the Barnes maze test, Akain1 KO mice and wild-type (WT) mice learned the fixed escape box position at a comparable rate. For reversal learning, in which the target was moved to the opposite side of the maze, the Akain1 KO mice and WT mice learned the new escape box position at a similar rate. While WT mice spent significantly more time investigating the new target than the original target, Akain1 KO mice exhibited no significant difference, suggesting that Akain1 KO mice had behavioral flexibility deficits. Together, these findings indicate that Akain1 is translated in the brain and has a critical role in the discrimination of similar environmental contexts.

Disclosures: K. Fujii: None. **Y. Koshidaka:** None. **M. Adachi:** None. **Y. Yanagibashi:** None. **M. Matsuo:** None. **S. Hongo:** None. **H. Nishizono:** None. **I. Takasaki:** None. **N. Kurosawa:** None. **Y. Aizawa:** None. **K. Takao:** None.

Poster

PSTR176. Animal Navigation

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

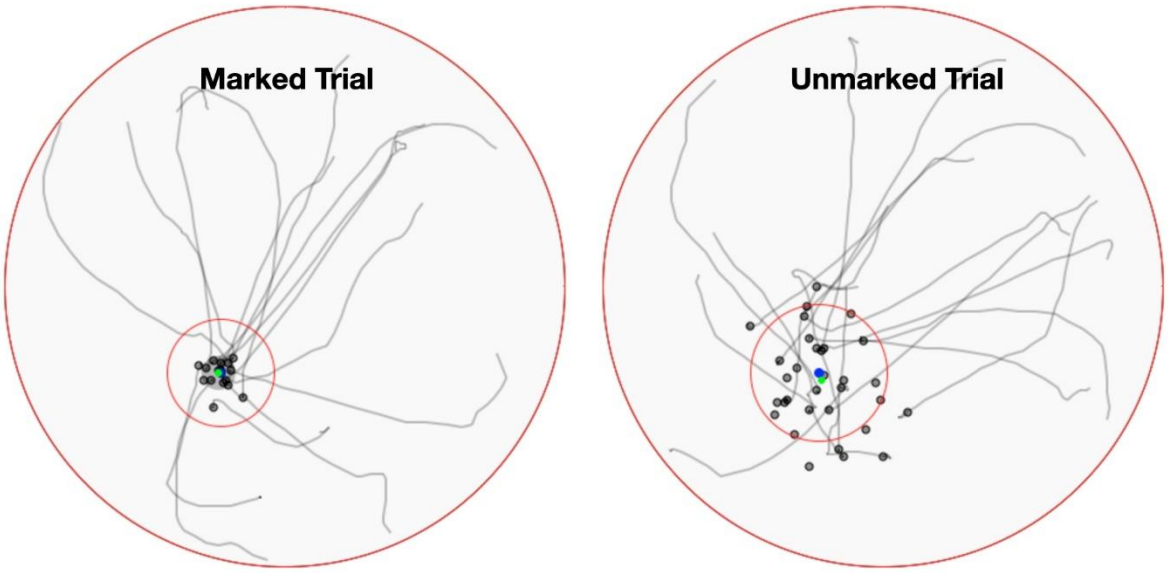
Program #/Poster #: PSTR176.09/TT23

Topic: H.09. Spatial Navigation

Title: Analysis of rat navigation

Authors: *J. KUBIE[1], B. M. DENVIR[2];
[1]SUNY Downstate Med. Ctr., Brooklyn, NY; [2]Intrnl. Med., NYU, New York, NY

Abstract: We developed an open-field task to measure rat navigation — paths and self-localization are analyzed. Each measure shows both accuracy and errors.
The task is modified from Kubie et al, 2007, where, in an open enclosure, a rat is rewarded for making a decision — pausing — near a goal location. Trials can be **"marked"** or **"unmarked"** based on whether there is an object in the center of the goal region**.** If the decision is accurate a food pellet is dropped to a random location. The critical modification in the current study was expanding the apparatus from 3ft to 6ft diameter. The gray circular chamber is gray and has a white cue card covering 90° of the wall.
Eight rats were trained using shaping procedures. All acheived asymptotic performance in about 40 days. Typically, in a 5-minute session a rat will make a dozen paths towards the goal followed by pause decisions. Paths and decisions are detected with an overhead camera and computer analyzed. A rat has a new goal area each day; the first trial of the day is marked, followed by 2 unmarked trials with the goal in the same location.
Self-localization accuracy was measured from pause locations. Rats were extremely accurate on the marked trials. On unmarked trials a rat's behavior has a shot-gun-scatter pattern centered on the goal. On a typical unmarked trial with a 9-inch reward-zone radius about 60% of the pauses will be in the reward zone. **Navitation** speed, linearity and accuracy were measured along a rat's path in a 2.5 sec time window prior to a pause. Typical paths were 5 to 10° deviant from goal, with length 110% of the shortest distance. Examples below:
Our conclusion is that the task is effective. Both self-localzation and navigation are efficient, wth equivalent accuracy when the goal is moved each day compared with static goals. Errors suggests a mismatch between veridical space and the rat's internal representation of space. Such 'errors' should be incorporated into place-cell analysis, and suggest re-interpretation of various results. The task is well suited to be used in conjunction with neuronal recording. Kubie et. al. (2007) Behavioral Neuroscience:121 (4)



Marked Trials have a disk in the goal center. **Unmarked trials** have no marker. "Choices" are dots. Choices in small circle are rewarded. Lines are three second paths to a choice.

Disclosures: **J. Kubie:** None. **B.M. Denvir:** None.

Poster

PSTR176. Animal Navigation

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR176.10/TT24

Topic: H.09. Spatial Navigation

Support: 1K99ES033256-01A1

Title: Sex differences in cognition from perinatal exposure to organophosphate flame-retardants in male and female adult offspring

Authors: *K. WIERSIELIS, R. MUKADAM, N. KNOX, T. DEGROAT, A. YASREBI, T. ROEPKE;
Rutgers Univ., New Brunswick, NJ

Abstract: Endocrine disrupting compounds (EDCs) are compounds found in our environment that interrupt typical endocrine function. A particular group of EDCs are flame-retardants due to their interaction with steroid and nuclear receptors. Humans are consistently exposed to flame-retardants daily as they are used in everyday items such as plastics, clothing, toys, and electronics. A commonly used class of flame retardants are the organophosphate flame-retardants (OPFRs), which have been shown to alter adult behavior in multiple rodent species after developmental exposures. We have previously identified that males perinatally exposed to OPFRs had a trending reduction in locomotion and females had a significant increase when in

the open field task. In addition, we detected OPFR-exposed males exhibited anxiolytic-like behavior in the elevated plus maze, although females had no difference. However, the effects of perinatal OPFR exposure on cognition and memory in the adult offspring are underexplored. Here we evaluate cognitive behavior using the Y-maze, spatial object recognition (SOR), novel object recognition (NOR), and the Barnes maze in male and female adult offspring that were perinatally exposed to a mixture of OPFRs (tris(1,3-dichloro-2-propyl)phosphate, triphenyl phosphate, tricresyl phosphate) or vehicle. In the Y-maze, we found OPFR-exposed males exhibited a reduction in the number of entries to the unknown arm and OPFR-exposed females exhibited an increase in comparison to same-sex controls, suggesting an impairment and enhancement in memory, respectively. In the SOR, a similar pattern emerged such that OPFR males exhibited a decrease in time spent with the displaced object and OPFR females showed an increase in contrast to controls. In both the NOR and the Barnes maze, we did not observe any treatment or sex differences. Our results demonstrate sex- and exposure-dependent effects of perinatal OPFR exposure on cognition in male and female adult mice, suggesting developmental neurotoxicity. Future studies will examine the influence of perinatal OPFR exposure on the long-term potentiation of pyramidal cells in hippocampal CA1 and acetylcholine excitatory synaptic transmission.

Disclosures: K. Wiersielis: None. **R. Mukadam:** None. **N. Knox:** None. **T. Degroat:** None. **A. Yasrebi:** None. **T. Roepke:** None.

Poster

PSTR176. Animal Navigation

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR176.11/TT25

Topic: H.09. Spatial Navigation

Support: NSERC RGPIN 2018-04060 LP
NSERC RGPIN 2017-4944 DK

Title: Does age and cognitive ability affect volume of the hippocampal formation in pigeons (Columba livia)?

Authors: *L. S. PHILLMORE[1], B. M. PARKS[1], S. LUPI[2], C. HAYAR[1], D. M. KELLY[3]; [1]Psychology and Neurosci., Dalhousie Univ., Halifax, NS, Canada; [2]Advanced Facility for Avian Res., Univ. of Western Ontario, London, ON, Canada; [3]Univ. of Manitoba, Univ. of Manitoba, Winnipeg, MB, Canada

Abstract: Pigeons are spatial specialists as they are able to navigate over long distances from new locations to their home roost. In parallel, they have a specialized neural region, the hippocampal formation (HF), that underlies this behaviour; adult pigeons with HF lesions can take up a homeward direction but cannot efficiently use landmarks to navigate accurately to a familiar location. Cognitive abilities appear to decline with age in homing pigeons but evidence

for age-related change in the HF has been relatively unclear, and the methodology used to estimate volume was rudimentary. To investigate more precisely how the brain, and specifically the HF might change with age, we collected brains from thirty-five male and female pigeons that were part of a colony and used in laboratory-based cognitive studies and ranged in age from 4-5 years to over 20 years. These pigeons had been subjects in various laboratory-based cognitive experiments including spatial memory. In a sequential learning task, the older pigeons had reduced memory capacity compared to younger pigeons, but this was not true of all older pigeons. By examining the brains of these birds, we can determine if there are general differences between younger and older pigeons, and individual differences among older pigeons that match the cognitive abilities measured in the sequential task. We stained brain tissue with cresyl violet and used the well-established histological technique of volume reconstruction using area measurements to estimate the absolute and relative volumes of the HF between adult (< 5 years), aged, and geriatric (>15 years) pigeons of both sexes and correlate this with our data on the sequential tasks. This study will provide further insight into the relationship between age and changes in HF of pigeons, and the relationship between cognitive capacity and HF volume as pigeons age.

Disclosures: L.S. Phillmore: None. **B.M. Parks:** None. **S. Lupi:** None. **C. Hayar:** None. **D.M. Kelly:** None.

Poster

PSTR176. Animal Navigation

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR176.12/TT26

Topic: H.09. Spatial Navigation

Support: Department of Defense

Title: Training and recall on the Lashley III maze, a low-stress route learning task, in male and female mice.

Authors: *L. B. TUCKER[1], A. A. GRILLAKIS[2], J. T. MCCABE[2];
[1]Preclinical Behavior and Modeling Core, [2]Anatomy, Physiology, And Genet., Uniformed Servies Univ. of the Hlth. Sci., Bethesda, MD

Abstract: The Lashley III Maze is a low-stress task that can be used to test egocentric route learning and memory. In contrast to other popular learning and memory tests like the Morris Water Maze and the Barnes Maze, the Lashley III Maze does not require food deprivation or aversive stimuli (such as bright overhead illumination) to motivate mice to navigate the test. Further, because the number of errors is measured in addition to the latency to find the goal, performance can be evaluated in animals with impaired motor function. This makes the Lashley III Maze appropriate for use with aging or injured animals. The maze consists of a start box and 4-arm body that must be navigated by test subjects to reach a goal box containing bedding from

the home cage. Openings in each arm of the maze require mice to make a decision whether to turn left or right, which leads to either progress through the maze or a dead-end. The total number of errors was measured for each mouse, including entry into a dead-end and backwards movement through the maze. C57Bl/6J 8-week-old male and female mice were trained on the maze for 5 days, with 2 trials per day, and then tested for recall 7 days later. The maze orientation was then reversed and mice were re-trained using the new orientation for an additional 5 days. The results of this pilot show that the number of errors made during navigation of the maze decreased, and that the number of errors remained low at the 7-day recall. Minimal errors were made during the reversal period. Both male and female C57Bl/6J mice are equally capable of route learning in the Lashley III Maze, and retain this learning up to one week after the training period. Additionally, mice are capable of re-learning a new maze orientation. This data suggests the Lashley III Maze is appropriate for use with C57Bl/6J mice, mice can be trained to navigate the maze successfully within 4-5 days, and are able to recall and re-learn new routes to the goal box.

Disclosures: L.B. Tucker: None. **A.A. Grillakis:** None. **J.T. McCabe:** None.

Poster

PSTR176. Animal Navigation

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR176.13/TT27

Topic: H.09. Spatial Navigation

Support: NRF Grant 2021R1A2C3005560
KIST institutional program 2E32211

Title: Stochastic characterization of navigation strategies in an automated variant of the Barnes maze

Authors: J. LEE, D. JUNG, ***S. ROYER**;
Korea Inst. of Sci. and Technol., Seoul, Korea, Republic of

Abstract: Animals can use a repertoire of strategies to navigate in an environment, and it remains an intriguing question how these strategies are selected based on the nature and familiarity of environments. To investigate this question, we developed a fully automated variant of the Barnes maze, characterized by 24 vestibules distributed along the periphery of a circular arena, and monitored the trajectories of mice over 15 days as they learned to navigate from a random start vestibule to a goal vestibule. We show that the patterns of vestibule visits can be reproduced by the combination of three stochastic processes reminiscent of random, serial and spatial strategies. The processes randomly selected vestibules based on either uniform (random) or biased (serial and spatial) probability distributions; closely matched experimental data across a range of statistical distributions characterizing the length, distribution, step size, direction, and stereotypy of vestibule sequences; and revealed a shift from random to spatial and serial

strategies over time, with a strategy switch occurring approximately every 6 vestibule visits. Our study provides a novel apparatus and analysis toolset for tracking the repertoire of navigation strategies and demonstrates that a set of stochastic processes can largely account for exploration patterns in the Barnes maze.

Disclosures: J. Lee: None. **D. Jung:** None. **S. Royer:** None.

Poster

PSTR176. Animal Navigation

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR176.14

Topic: H.09. Spatial Navigation

Title: Navigational strategy selection in a dual solution plus-maze

Authors: *L. ANDREOLI[1], S. TOMONAGA[1], A. C. A. SALMERON[1], J. R. WICKENS[2], K. TANAKA[1];
[1]Okinawa Inst. of Sci. and Technol., Onna, Japan; [2]Neurobio. Res. Unit, Okinawa Institue of Sci. and Technol., Onna, Japan

Abstract: Whether it is to find food, shelter, or a mate, navigation is a behaviour extremely important for all animals. Finding a goal location can be done by using either allocentric cues from the environment or egocentric cues from the animal's own body and self-motion. The dual solution plus-maze is a behavioural task designed to probe whether the animal is using allocentric (place strategy) or egocentric (response strategy) cues when navigating towards a goal. Classic studies using this maze have demonstrated that animals rely initially on place strategy and, after navigating several times in the same environment, start utilising response strategy. The aim of our study was to assess if availability of allocentric cues could influence the use of navigational strategy, as well as provide a different approach to evaluate suitability of the environment for the dual solution plus-maze task. For that, we trained mice in a dual solution-plus maze and in an adapted version of the maze where we forced animals to use one of the strategies. We measured the number of days for animals to reach our learning criteria (70% accuracy in two consecutive days) and compared the number of animals that used each type of strategy during a probe trial. In our study animals significantly used response strategy more often than place strategy, even during early training and in an environment where several allocentric cues were available. We also showed that the availability of allocentric cues influenced how well the animals learn each strategy, regardless of their preference for response strategy when being probed. Our results make us question the prevailing view that when navigating in the dual solution plus-maze, animals initially rely on allocentric place strategy and after extensive training shift to egocentric response strategy. More important than the time of training, we propose that characteristics of the environment (and possibly the complexity of the navigational behaviour) play a key role in animal's selection of navigational strategies.

Disclosures: **L. Andreoli:** None. **S. Tomonaga:** None. **A.C.A. Salmeron:** None. **J.R. Wickens:** None. **K. Tanaka:** None.

Poster

PSTR176. Animal Navigation

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR176.15/TT28

Topic: H.09. Spatial Navigation

Title: The Role of the Medial Prefrontal Cortex in Memory and Spatial Navigation in the Traveling Salesperson Problem

Authors: *J. SPAULDING[1], M. TAVAREZ[1], G. KOBZEFF[1], A. SALAZAR[2], R. BLASER[1], J. B. HALES[1];
[1]Univ. of San Diego, San Diego, CA; [2]Univ. of Chicago, Chicago, IL

Abstract: Cognitive impairment is a devastating feature of many neurological conditions, including Alzheimer's disease, traumatic brain injury, and stroke. Some of the most impactful deficits involve memory loss, disrupted spatial navigation, and poor executive function. Effective research into the causes and treatments of such disorders requires knowledge of the neurological mechanisms that underlie these processes. The Traveling Salesperson Problem (TSP) is an optimization task that requires subjects to identify the shortest route to travel from a starting to ending point, while visiting a certain number of targets. This task has been used to examine spatial memory and decision making in animal models, such as rats, while they perform naturalistic foraging behaviors. Previous studies from our lab have found that rats with hippocampal lesions and medial entorhinal cortex lesions are impaired across many different measures and spatial configurations in the TSP task, specifically on measures of spatial memory but not spatial decision making. Given the various cognitive demands of this task, our lab was interested in examining whether the medial prefrontal cortex (mPFC) was responsible for spatial decision making in the TSP. We tested both male and female rats on the TSP task following excitotoxic lesions of the mPFC or sham lesions. Behavioral analyses examined the effects of lesion, sex, navigational strategy, and spatial configurations of the targets on various measures of spatial memory and decision making. Preliminary results found significant interactions between lesion and navigational strategy, lesion and spatial configuration of the targets, and lesion and sex across multiple measures, suggesting a complex role of the mPFC in both spatial memory and decision making.

Disclosures: **J. Spaulding:** None. **M. Tavarez:** None. **G. Kobzeff:** None. **A. Salazar:** None. **R. Blaser:** None. **J.B. Hales:** None.

Poster

PSTR176. Animal Navigation

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR176.16/UU1

Topic: H.09. Spatial Navigation

Support: UCSF Discovery Fellows Program
HHMI 2014-146122-2014359-45

Title: A locomotor rhythm organizes directional firing of neurons in the superior colliculus

Authors: *C. WILHITE[1], L. M. FRANK[2,3,4], M. SCANZIANI[5,3,4];
[1]Neurosci. Grad. Program, [2]Physiol. and Psychiatry, UCSF, SAN FRANCISCO, CA; [3]Howard Hughes Med. Inst., Chevy Chase, MD; [4]Kavli Inst. for Fundamental Neurosci., San Francisco, CA; [5]Physiol., Univ. of California, San Francisco, San Francisco, CA

Abstract: Studies investigating the role of the superior colliculus (SC) in spatial orienting behavior have traditionally involved partially restrained animals performing orienting movements toward external stimuli. Accordingly, how neural activity in SC is organized during freely moving, internally driven behaviors is unknown. Here we record SC neurons in freely moving mice during a Y-maze spatial foraging task and identify neurons that preferentially fire during either left or right turns at the bifurcation of the maze. Remarkably, we show that a subset of these neurons fire rhythmically, in phase with a ~6 Hz left/right head oscillation that is characteristic of locomotion. Strikingly, a neuron's turn direction preference on the Y-maze (i.e., selective firing during left or right turns) largely determines its preferred firing phase relative to the locomotor rhythm. During this preferred firing phase, the preferred turn direction of SC neurons matches the direction of the ongoing head oscillation. As a result, rhythmic neurons in SC that prefer opposite turn directions are segregated into two populations that fire during opposite phases/directions of the locomotor head rhythm. In summary, neural activity in mouse SC involves distinct populations whose firing alternates such as to represent rhythmic directional movements of the animal as it locomotes through space.

Disclosures: C. Wilhite: None. **L.M. Frank:** None. **M. Scanziani:** None.

Poster

PSTR176. Animal Navigation

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR176.17/UU2

Topic: H.09. Spatial Navigation

Support: HHMI
NIH Grant U19

Title: Inter- and intra-hemispheric sources of vestibular signals to the primary visual cortex

Authors: *G. BOUVIER[1,2,3], A. SANZENI[4,5], E. HAMADA[1], N. BRUNEL[5], M. SCANZIANI[1,3];
[1]Univ. of California, San Francsico, San Francisco, CA; [2]Paris-Saclay Inst. of Neurosci., Saclay, France; [3]Howard Hughes Med. Inst., San Franscico, CA; [4]Bocconi Univ., Milan, Italy; [5]Duke Univ., Durham, NC

Abstract: Head movements are sensed by the vestibular organs. Unlike classical senses, signals from vestibular organs are not conveyed to a dedicated cortical area but are broadcast throughout the cortex. Surprisingly, the routes taken by vestibular signals to reach the cortex are still largely uncharted. Here we show that the primary visual cortex (V1), a cortical area where head movement variables, including direction, velocity and acceleration, are accurately encoded, receives these signals from the ipsilateral pulvinar and the contralateral visual cortex. The ipsilateral pulvinar provides the main head movement signal, with a bias toward contraversive movements (e.g. clockwise movements relative to left V1). Conversely, the contralateral visual cortex provides head-movement signals during ipsiversive movements. Crucially, head movement variables encoded in V1 are already encoded in the pulvinar, suggesting that those variables are computed subcortically. Thus, the convergence of inter and intra-hemispheric signals endows V1 with a rich representation of the animal's head movements.

Disclosures: G. Bouvier: None. **A. Sanzeni:** None. **E. Hamada:** None. **N. Brunel:** None. **M. Scanziani:** None.

Poster

PSTR176. Animal Navigation

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR176.18

Topic: H.09. Spatial Navigation

Title: The Impact of the Tecto-Pulvinar Pathway on Spatial Representations in the Hippocampus

Authors: *J. M. BRENNER[1], S. RUEDIGER[2], R. BELTRAMO[3], M. SCANZIANI[4];
[1]Baylor Col. of Med., Houston, TX; [2]Univ. Col. London, London, United Kingdom; [3]Univ. of Cambridge, Cambridge, United Kingdom; [4]Univ. of California, San Francisco, San Francisco, CA

Abstract: The postrhinal cortex (POR) is considered one of the main gateways of visual information to the hippocampus. We have recently shown that POR receives visual input from the superior colliculus via the tecto-pulvinar pathway (Brenner et al. 2023). What is the impact of this pathway on spatial representation in the hippocampus? We trained animals to traverse a linear track for reward. Recording from neurons in CA1 of the hippocampus revealed place field maps that tile the entire length of the linear track. Switching from light to darkness (as defined by the number of retinal photoisomerizations) led to a drastic remapping of the spatial organization

of place fields without any impact on the behavior of the animal. The original spatial map was reinstated upon returning to light. Silencing the tecto-pulvinar pathway with the conditional expression of tetanus toxin light chain abolishes the remapping; that is, the maps in light are identical to the maps in darkness. Thus, the tecto-pulvinar pathway contributes visual information used for the representation of space in the hippocampus.

Disclosures: **J.M. Brenner:** None. **S. Ruediger:** None. **R. Beltramo:** None. **M. Scanziani:** None.

Poster

PSTR176. Animal Navigation

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR176.19/UU3

Topic: H.09. Spatial Navigation

Support: NIH Grant NS053907

Title: Is the Head-Direction System sensitive to three-dimensional tuning?

Authors: *Z. HAGBI, R. M. GRIEVES, J. S. TAUBE;
Psycological and Brain Sci., Dartmouth Col., Hanover, NH

Abstract: While orienting in an environment, animals use several types of neurons in various brain regions to encode different aspects of spatial orientation. One prominent variable is directional heading, believed to be encoded by cells in the head direction (HD) network, which consists of cells that fire when the animal's head is pointing in a specific direction in the horizontal (azimuthal) plane. However, the real world is three-dimensional, and even surface-bounded terrestrial animals often encounter various three-dimensional structures while orienting and navigating for basic life needs. This situation raises the question of how neurons represent three-dimensional space in the brain, and more specifically, what are the three-dimensional properties of HD cells. Some studies have contended that HD cells encode three-dimensional space, while other studies have argued against this notion. In this study, we focused on the question of whether HD cells are sensitive to the tilt of the animal's head. To address this issue, we designed an apparatus that was shaped as a hilly-terrain that enabled us to monitor rat HD cells from the anterodorsal thalamus in a laboratory setting. Locomotion on the hill-maze encourages the rats to travel in different directions that promote sampling of different tilt orientations of the head. Since rats are surface-bounded animals and travel most of the time with their head aligned to the horizontal plane, we hypothesize that HD cell activity will be mainly modulated by the horizontal plane of the environment and not the degree of tilt of the rat's head. Indeed, our data indicate that thalamic HD cells are not modulated by the rat's head tilt orientation, even in a three-dimensional environment. This result is consistent with the view that HD cells, at least in the anterior thalamus, encode the rat's HD with respect to its current plane of locomotion.

Disclosures: **Z. Hagbi:** None. **R.M. Grieves:** None. **J.S. Taube:** None.

Poster

PSTR176. Animal Navigation

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR176.20/UU4

Topic: H.09. Spatial Navigation

Support: NIH Grant NS053907

Title: Hippocampal place cells utilize terrain shape for spatial localization

Authors: *R. M. GRIEVES, E. DUVELLE, J. S. TAUBE;
Psychological and Brain Sci., Dartmouth Col., Hanover, NH

Abstract: Research suggests that we construct an internal spatial representation of our environment, or a 'cognitive map', and use this for spatial navigation (O'Keefe and Nadel, 1978). Three main cell types have been implicated in these processes: 1) hippocampal place cells which represent specific 'places' in an environment; 2) grid cells which map space with a continuous hexagonal pattern of firing fields and 3) head direction cells which represent the orientation of the animal's head. We know a great deal about how these spatial neurons represent flat, horizontal surfaces from research spanning decades. More recently, studies investigating the third dimension of navigation found that place cells represent space volumetrically and use identical mapping rules in two- and three-dimensions (Grieves et al. 2020). However, surface-bound animals such as rats and humans spend most of their time navigating across uneven surfaces where terrain shape can complicate navigation but also support it by providing additional localizing cues. Thus, it would be advantageous for a cognitive map to include this information. To test if place cells are purely volumetric or represent informative terrain cues, we implanted rats with electrodes in the dorsal hippocampus (CA1). We then recorded the activity of place cells in a large (3 m × 1.5 m) horizontal arena or a region of hilly terrain. We found that place cell activity was indeed affected by the local terrain: 1) place fields were often elongated parallel to slopes; 2) despite this response, place fields were smaller and more precise in the hilly terrain than the horizontal arena; and 3) many cells exhibited multiple firing fields that occurred in locations with similar local terrain features. Together, these results suggest that place cells do not map space in a purely volumetric manner, instead they represent the shape of local terrain. Additionally, terrain affects place cells in a similar way to environment geometry (i.e., walls), suggesting a possible role for boundary selective neurons. Future experiments will look to determine how the activity of boundary, grid and head direction cells support this three-dimensional cognitive map.

Disclosures: **R.M. Grieves:** None. **E. Duvelle:** None. **J.S. Taube:** None.

Poster

PSTR176. Animal Navigation

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR176.21/UU5

Topic: H.09. Spatial Navigation

Support: NIH grant NS053907
NS111695

Title: Angular head velocity cells within brainstem nuclei projecting to the head direction circuit

Authors: *J. GRAHAM[1], J. R. DUMONT[5], S. S. WINTER[2], J. E. BROWN[3], P. A. LACHANCE[4], C. C. AMON[1], K. B. FARNES[1], A. J. MORRIS[1], N. STRELTZOV[1], J. S. TAUBE[6];
[1]Dartmouth Col., Hanover, NH; [2]Home, Dartmouth Col., Lebanon, NH; [3]Dept Ophthalmol & Visual Sci., [4]Psychological and Brain Sci., Dartmouth Col., Hanover, NH; [5]Psychological & Brain Sci., Dartmouth Col., Hanover, NH; [6]Dartmouth Col., Darmouth Col., Hanover, NH

Abstract: An animal's perceived sense of orientation depends upon the head direction (HD) system found in several limbic structures which, in turn, depends upon an intact peripheral vestibular labyrinth. However, how the vestibular system influences the generation, maintenance and updating of the HD signal remains poorly understood. Anatomical and lesion studies point towards three key brainstem nuclei as being potential critical components in generating the HD signal: nucleus prepositus hypoglossi (NPH), supragenual nucleus (SGN), and dorsal paragigantocellularis reticular nuclei (PGRNd). Collectively, these nuclei are situated between the vestibular nuclei and the dorsal tegmental and lateral mammillary nuclei, which are thought to serve as the origin of the HD signal. Here, we record from each of these three brain areas in either freely-moving or passively rotated rats. During free foraging, two fundamental types of AHV cells were observed: 1) symmetrical AHV cells increased or decreased their neural firing with increases in AHV regardless of the direction of rotation; 2) asymmetrical AHV cells responded differentially to clockwise (CW) and counter-clockwise (CCW) head rotations. When rats were passively rotated, some AHV cells remained sensitive to AHV whereas others had attenuated firing. In addition, many AHV cells were modulated by linear head velocity. These results indicate the types of information conveyed in the ascending vestibular pathways that could be responsible for generating the HD signal.

Disclosures: J. Graham: None. **J.R. Dumont:** None. **S.S. Winter:** None. **J.E. Brown:** None. **P.A. Lachance:** None. **C.C. Amon:** None. **K.B. Farnes:** None. **A.J. Morris:** None. **N. Streltzov:** None. **J.S. Taube:** None.

Poster

PSTR176. Animal Navigation

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR176.22/UU6

Topic: H.09. Spatial Navigation

Support: CRC T2 in Neural Circuits of Cognition and Control
CFI-JELF
UBC Four Year Doctoral Fellowship

Title: Examining hippocampal task-relevant coding in rats freely moving in a virtual-reality Dome apparatus

Authors: *W. FANG[1], R. KORNELSEN[2], A. DHIR[3], I. MORGAN[3], M. MADHAV[3]; [1]Grad. Program in Neurosci., [2]Djavad Mowafaghian Ctr. for Brain Hlth., [3]Sch. of Biomed. Engin., Univ. of British Columbia, Vancouver, BC, Canada

Abstract: The hippocampal formation is thought to encode the cognitive map, classically defined as a metric Euclidean representation of the spatial location of an animal. Recent studies indicate that hippocampal neurons can additionally represent non-spatial domains informed by cues that are informative of the animal's behavioral task. We investigate hippocampal encoding of task-relevant cues in freely moving rodents using a virtual-reality Dome apparatus. In the Dome, rats run freely on a circular track while being surrounded by projected visual cues and listening to auditory cues. Rats are trained to request reward through nose-pokes at specific locations within the task-relevant auditory reference frame, which moves with respect to the stationary lab frame according to an auditory gain value. Projected landmarks form a visual reference frame that can be similarly moved according to a visual gain value. Behavioral data shows that rats were able to perform this auditory location task with high accuracy. We recorded population activity in hippocampal CA1, and quantified spatial information scores of CA1 place cells in the circular task-relevant (auditory) and task-irrelevant (visual) frames, and a conjunctive toroidal frame. Preliminary data indicates that CA1 neurons encode locations in the conjunctive space formed by the sound and the visual frames. We will test whether the relative information content across the three reference frames (lab, visual and auditory) is dependent on their task relevance. We will also independently quantify the topology of the neural representation using geometric deep learning techniques and compare it with the topology of the task.

Disclosures: W. Fang: None. **R. Kornelsen:** None. **A. Dhir:** None. **I. Morgan:** None. **M. Madhav:** None.

Poster

PSTR176. Animal Navigation

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR176.23/UU7

Topic: H.09. Spatial Navigation

Support: UBC SBME faculty startup funds
T2 CRC in Neural Circuits of Cognition and Control
CFI – John R. Evans Leaders' Fund
Djawad Mowafaghian Centre for Brain Health Alzheimer's Disease Research Grant
Jack Brown and Family Alzheimer Research Foundation Grant

Title: The Omniroute maze: dynamically configurable routes and sensory cues to investigate rodent navigation

Authors: *A. W. LESTER, A. G. MOMBEINI, N. N. DJAFRI, A. DHIR, M. NARIMANI, G. KAUR, M. S. MADHAV;
Sch. of Biomed. Engin., Univ. of British Columbia, Vancouver, BC, Canada

Abstract: We developed a novel physical maze that enables highly flexible real-time experimental control over available routes and cues, comparable to that afforded by virtual reality (VR) systems but in the context of unconstrained real-world rodent behaviour. The maze is composed of a 1.5 x 1.5 m platform with 113 independently movable wall segments that can be programmatically activated to generate unique routes within a 5 x 5 grid. The use of octagonal geometry accommodates both orthogonal and diagonal path trajectories. Four projectors arrayed around the perimeter of the maze can display distinct visual cues on either side of any subset of raised walls. The system also incorporates high-speed 3D tracking of rat position and orientation for closed-loop control of the available paths based on real-time behavior. Additionally, an automated gantry system provides food-based reinforcement anywhere in the maze. These hardware components and the electrophysiological data collection system are controlled using the Robot Operating System (ROS) framework. The Omniroute maze can reproduce classic behavioural mazes such as the Y-maze, T-maze and W-maze, along with a variety of other configurations to test hypotheses regarding the flow of information between environmental configurations, cues, neural representations and navigational decisions. The automated reconfiguration, tracking and reward delivery supports high-throughput behavioral assays of complex navigation behavior without the potential confounds resulting from direct experimenter intervention. The Omniroute maze was designed from the ground up for robust operation using affordable, non-proprietary, and open-source components, hardware and software, with an eye toward accessible fabrication and assembly that will aid replicability by other investigators.

Disclosures: **A.W. Lester:** None. **A.G. Mombeini:** None. **N.N. Djafri:** None. **A. Dhir:** None. **M. Narimani:** None. **G. Kaur:** None. **M.S. Madhav:** None.

Poster

PSTR176. Animal Navigation

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR176.24/UU8

Topic: A.07. Developmental Disorders

Support: TRiO McNair

Title: Naturalistic spatial memory and decision making in a rodent model of attention deficit hyperactivity disorder (ADHD)

Authors: Z. WYNTER, T. DUQUE, M. WYATT, A. BECKETT, *J. B. HALES;
Univ. of San Diego, San Diego, CA

Abstract: In order to successfully navigate in our environment, we rely on the critical ability to remember spatial locations of objects and to make navigational decisions based on that information. Without this ability, we would get lost and forget where we placed items. Unfortunately, these tasks can be considerably more difficult for individuals with certain neurological conditions, such as Alzheimer's disease and attention deficit hyperactivity disorder (ADHD). Our lab has used a task known as the Traveling Salesperson Problem (TSP) to examine the complex cognitive processes involved in naturalistic spatial foraging in rats. The TSP is an optimization task that requires subjects to identify the shortest route to travel from a starting to ending point while visiting a certain number of targets in an open arena. This spontaneous behavior involves multiple cognitive processes, including spatial working memory, spatial orientation, and executive processing. Previous research from our lab has found that rats with damage to the hippocampus show deficits in measures of spatial memory on the TSP. Interestingly, people with ADHD have shown similar deficits in spatial working memory. ADHD is a neurological disorder characterized by hyperactivity, impulsivity, and inattention, as well as deficits in both working memory and sense of time. Although animal models cannot fully reflect human neurological conditions, they can provide insight into the disorder that cannot be obtained from human studies. Our study examined the performance of Spontaneously Hypertensive Rats (SHR), the most widely used rodent model of ADHD, relative to their control model, Wistar Kyoto (WKY) rats, on the TSP task. We used male and female rats because, in both rodent and human studies of ADHD, females have been understudied, and there is a critical need to examine sex as a biological variable. Preliminary results found a significant interaction between sex and strain in the latency for rats to retrieve all targets. In addition, the number of revisits per target (a measure of spatial working memory errors) increased as the number of total targets increased, with a marginally significant effect of sex. Our preliminary findings suggest both sex and strain differences in TSP performance on measures related to spatial working memory and navigational strategies.

Disclosures: Z. Wynter: None. **T. Duque:** None. **M. Wyatt:** None. **A. Beckett:** None. **J.B. Hales:** None.

Poster

PSTR177. Neuropathology, Genetics, Genomics, and Molecular Mechanisms: Schizophrenia

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR177.01/UU9

Topic: H.13. Schizophrenia

Support: NIH Grant R01MH080234

Title: Genomic and transcriptomic profiles underlying the effect of Setd1a disruption on mouse cortical excitatory neurons

Authors: *P. APOSTOLOU[1,2], Z. SUN[2,3], Y. CHEN[1], B. XU[3], J. GOGOS[1,2,3,4];
[1]Mortimer B. Zuckerman Mind Brain and Behavior Inst. Columbia Univ., New York, NY;
[2]Dept. of Physiol. and Cell. Biophysics, [3]Dept. of Psychiatry, [4]Dept. of Neurosci., Columbia Univ., New York, NY

Abstract: Rare de novo or inherited genetic variants have been identified as high-risk factors for schizophrenia (SCZ) through extensive sequencing studies. We have previously established a link between loss-of-function *de novo* mutations in SETD1A and SCZ risk, a finding confirmed by large-scale exome sequencing studies, which identified SETD1A as the risk gene with the largest statistical support for SCZ. SETD1A encodes a lysine methyltransferase best known for its role in the Set/COMPASS complex, mediating methylation of lysine 4 on histone H3 and regulating gene expression. We have previously shown that mice carrying a heterozygous loss-of-function mutation of the orthologous gene exhibit morphological and functional alterations in cortical excitatory neurons accompanied by cognitive deficits. To identify genomic and transcriptomic profiles underlying these effects we performed Cut&Tag sequencing in excitatory neurons purified from brains of wild type and *Setd1a* mutant mice using fluorescence- activated cell sorting. By integrating these data with bulk RNA sequencing in the same neurons, we established a connection between Setd1a binding and the corresponding gene expression patterns. Chromatin profiling combined with RNA sequencing revealed a large number of Setd1a binding sites, distributed over regulatory elements in the mouse genome and allowed for parallel comparisons of gene expression and chromatin binding, leading to the identification of high-confidence target genes as well as potential Setd1a recruiters and interactors that mediate the effects of Setd1a on excitatory neurons in the adult brain. In ongoing experiments we investigate the impact of neuronal activity on the transcriptional effects of Setd1a deficiency by utilizing a chemogenetic approach to induce synchronous neuronal activity in mouse cortical pyramidal neurons. Our results provide valuable insights into the primary transcriptional alterations caused by SETD1A mutations and enhance our understanding of the underlying regulatory mechanisms that impact cortical pyramidal gene expression and their relevance to SCZ.

Disclosures: P. Apostolou: None. **Z. Sun:** None. **Y. Chen:** None. **B. Xu:** None. **J. Gogos:** None.

Poster

PSTR177. Neuropathology, Genetics, Genomics, and Molecular Mechanisms: Schizophrenia

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR177.02/UU10

Topic: H.13. Schizophrenia

Support: R21 MH128462

Title: Single-cell and spatial genomics of cerebellar cell type-specific molecular adaptations in schizophrenia and bipolar disorder

Authors: M. CORTES-GUTIERREZ[1], ***A. BISWAS**[1,2], B. HERB[1,3], R. JOHNSON[4], R. SCHWARCZ[4], S. AMENT[1,4];
[1]Inst. for Genome Sci., [2]Program in Mol. Med., [3]Dept. of Pharmacology,, [4]Maryland Psychiatric Res. Center, Dept. of Psychiatry, Univ. of Maryland Sch. of Med., Baltimore, MD

Abstract: Roles for the cerebellum in psychiatric disorders remain understudied, despite evidence for anatomical and functional differences in affected individuals. We sequenced the nuclear transcriptomes and chromatin accessibility states of 446,708 cells from the post-mortem cerebellum of individuals who died with schizophrenia (n=16), bipolar disorder (n=16), and unaffected controls (n=20). Samples were derived from posterior vermal regions of the cerebellar cortex. Donors included equal numbers of males and females and ranged from 16-68 years old at the time of death. We analyzed these data together with existing bulk cerebellum gene expression profiles from an independent case-control cohort (n=144). We found cell type-specific gene regulatory differences in donors who died with schizophrenia or bipolar disorder, including the down-regulation of certain genes expressed specifically at the synapses of two cerebellar neuron subtypes, Purkinje cells and granule cells. We are developing protocols for spatial transcriptomics with the Curio Biosciences (Slide-seq) platform to validate these findings and gain insight into their spatial organization within the cerebellar cortex. In summary, our study reveals associations of psychiatric disorders with molecular adaptations in cerebellar neurons.

Disclosures: M. Cortes-Gutierrez: None. **A. Biswas:** None. **B. Herb:** None. **R. Johnson:** None. **R. Schwarcz:** None. **S. Ament:** None.

Poster

PSTR177. Neuropathology, Genetics, Genomics, and Molecular Mechanisms: Schizophrenia

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR177.03/UU11

Topic: H.13. Schizophrenia

Support: The Rutgers-Princeton Center for Computational Cognitive Neuropsychiatry (CCNP) Pilot Grant

Title: Clinical and cognitive significance of buccal cell NOS1AP biomarkers for schizophrenia

Authors: *C. CROSTA[1], J. STUCKEY[1], A. BHATTIPROLU[1], J. SOLIS[2], B. L. FIRESTEIN[1]; [1]Cell Biol. and Neurosci., [2]Brain Hlth. Inst., Rutgers Univ., Piscataway, NJ

Abstract: Schizophrenia (SCZ) is a heterogenous and polygenic psychiatric illness characterized by the presence of positive, negative, and cognitive symptoms. Since little is known about the molecular and genetic etiology of the disease, current therapeutics are often inadequate for treating all associated symptoms. Commonly prescribed antipsychotics can alleviate positive symptoms of SCZ but do little to address the negative and cognitive symptoms, resulting in high rates of lifelong disability. Identification of disease-associated biomarkers would aid in the development of novel therapeutics and facilitate the implementation of biologically-driven diagnostic criteria. The N-methyl-D-aspartate receptor (NMDAR) hypofunctioning hypothesis suggests that symptoms of the disease are caused by a reduction in NMDAR signaling. Nitric oxide synthase 1 adaptor protein (NOS1AP) is encoded by a SCZ susceptibility gene, disrupts nitric oxide signaling, and negatively regulates NMDAR signaling. We previously reported that NOS1AP expression is increased in postmortem brain samples from patients with SCZ. Here, a cohort of patients with SCZ (n=37) and age-, race-, and gender-matched control subjects (n=30) were recruited to determine if NOS1AP SNPs, levels of NOS1AP mRNA, and levels of NOS1AP protein in human buccal cells could serve as biomarkers of SCZ. To characterize these possible biomarkers, we assessed their relationship with symptom severity and cognitive dysfunction. Preliminary analysis of our cognitive data determined that patients with SCZ have significantly increased reaction times and decreased response accuracy when compared to control subjects in the AX continuous performance (AX-CPT) task, suggesting deficits in cognitive control. Moreover, patients with SCZ also had deficits in response accuracy in the Jittered-Orientation Visual Integration (JOVI) task, suggesting deficits in visual integration. RT-qPCR, Western blot analysis, and a proteomics assay have been performed. Correlational analyses have been used to assess the relationship between these variables and cognitive performance. Cumulatively, this work has the potential to identify easy to collect, novel biomarkers of SCZ, thus providing insight into disease pathophysiology, improving diagnostic criteria, and diversifying therapeutic options.

Disclosures: C. Crosta: None. **J. Stuckey:** None. **A. Bhattiprolu:** None. **J. Solis:** None. **B.L. Firestein:** None.

Poster

PSTR177. Neuropathology, Genetics, Genomics, and Molecular Mechanisms: Schizophrenia

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR177.04/Web Only

Topic: H.13. Schizophrenia

Title: Regional cortical thinning in multiple-episode psychosis patients compared to first-episode psychosis patients and their association with clinical and cognitive features

Authors: *N. KANG, M. BANG, S.-H. LEE;
CHA bundang medical center, Sung-nam, Korea, Republic of

Abstract: Background: Schizophrenia is a devastating psychiatric disorder accompanied by abnormal brain structure, including cortical thickness, a sensitive measure for cytoarchitectural characteristics and alterations in cortical structures. Recurrent relapse of psychotic episode might deteriorate both the brain structure and corresponding neurocognitive function. Although previous studies revealed relationships between cortical thickness and corresponding executive function in fronto-temporal brain regions in schizophrenia patients, the effect of recurrent relapse of illness on these relationships remains unclear. Thus, we aimed to investigate the differences in cortical thickness between patients with first-episode psychosis (FEP) and multiple-episode psychosis (MEP) and explore their relationships with executive function.
Methods: One hundred twenty five patients with FEP (89 women) and 81 patients with MEP (53 women) underwent T1-weighted magnetic resonance imaging (MRI) scan. The regional cortical thickness difference in the cerebrum between two groups was examined using FreeSurfer. Executive function were measured using Wisconsin Card Sorting Test (WCST). All analyses were performed with age, sex, and intracranial volume as covariates.
Results: Compared to FEP participants, MEP participants had significantly lower cortical thicknesses in the left superior frontal gyrus ($p < 0.001$), left pars triangularis ($p = 0.001$), left isthmus cingulate gyrus ($p = 0.001$), left fusiform ($p = 0.003$), and right superior frontal gyrus ($p < 0.001$). Thinner right superior frontal gyrus was significantly associated with lower scores in conceptual level response in WCST ($r = 0.281$, $p = 0.009$).
Discussion: Our findings suggest that patients with MEP had decreased cortical thickness in frontal and temporal brain regions, which are previously known to be more affected than other regions in schizophrenia patients. As the number of relapses increases, cortical thickness in these regions is decreased, leading to poorer executive function. Since frequent relapses can have negative impact on the brain structures and executive functions, it is important to pay close attention from the early stages.

Disclosures: N. Kang: None. **M. Bang:** None. **S. Lee:** None.

Poster

PSTR177. Neuropathology, Genetics, Genomics, and Molecular Mechanisms: Schizophrenia

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR177.05/UU12

Topic: H.13. Schizophrenia

Support: NIH Grant (NIGMS r35GM13342)

Title: Uncovering the polypharmacology of clozapine reveals novel mechanisms of action

Authors: *J. K. LANHAM[1], A. B. CAO[2], J. D. MCCORVY[3];
[1]Cell Biol. Neurobio. and Anat. (CBNA), Med. Col. of Wisconsin, Milwaukee, WI; [3]Cell Biol. Neurobio. and Anat. (CBNA), [2]Med. Col. of Wisconsin, Wisconsin, WI

Abstract: Uncovering the polypharmacology of clozapine reveals novel mechanisms of action

Authors: Janelle K. Lanham[1], Andrew B. Cao[1], John D. McCorvy[1]
Department of Cell Biology, Neurobiology, and Anatomy, Medical College of Wisconsin

Atypical antipsychotics, such as clozapine and olanzapine, manage both positive and negative symptoms of schizophrenia via antagonism of dopamine (D2) and serotonin (5-HT_{2A}) receptors, respectively. However, their full polypharmacological profile is still understudied, which may contribute toward undiscovered additional therapeutic targets or side-effects. For example, clozapine is the preferred treatment in drug-resistant schizophrenia and shows efficacy in treating cognitive deficits associated with schizophrenia. However, clozapine has life-threatening side-effects including agranulocytosis. By contrast, olanzapine does not possess agranulocytosis side-effects, but is less effective in treating cognitive deficits. Considering the close structural similarity between clozapine and olanzapine, we sought to discover differences in their polypharmacological profile that would explain their differences in clinical efficacy. Therefore, we interrogated agonist and antagonist activity comparing clozapine to olanzapine at 33 aminergic G protein-coupled receptors (GPCRs) using a Bioluminescence Resonance Energy Transfer (BRET). Importantly, we also interrogated potential active *N*-desmethyl metabolites of clozapine and olanzapine and compared activities across several GPCRs to the non-dopaminergic/serotonergic antipsychotic, Xanomeline, which is in Phase 3 clinical trials for schizophrenia. We uncovered that clozapine, and not olanzapine, had unexpected potent agonist activity at other 5-HT receptor subtypes and at mAChR2 and mAChR4 muscarinic receptors, which are potential targets for treating cognitive deficits. Taken together, our results reveal that off-target activity other aminergic GPCR activity may explain the superior clinical efficacy for some atypical antipsychotics and this study reveals key GPCR targets to factor in future antipsychotic drug design.

Support: National Institutes of Health General Medical Sciences grant (NIGMS R35GM13342)

Disclosures: J.K. Lanham: None. **A.B. Cao:** None. **J.D. McCorvy:** None.

Poster

PSTR177. Neuropathology, Genetics, Genomics, and Molecular Mechanisms: Schizophrenia

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR177.06/UU13

Topic: H.13. Schizophrenia

Support: NIDA grant U01DA048279

Title: Cell specific transcriptome and 3D genome profiling in ventral midbrain of subjects with Schizophrenia and Bipolar Disorder

Authors: *S. SINGH[1], M. ISKHAKOVA[1], T. LAMBERT[1], S. MARENCO[6], P. AULUCK[6], G. HOFFMAN[1,2,3,4,5], P. ROUSSOS[1,2,3,4,5], K. GIRDHAR[1,2,3,4,5], S. AKBARIAN[1,3];
[1]Dept. of Psychiatry, [2]Ctr. for Dis. Neurogenomics, [3]Friedman Brain Inst., [4]Icahn Inst. for Data Sci. and Genomic Technol., [5]Dept. of Genet. and Genomic Sci., Icahn Sch. of Med. at Mount Sinai, New York, NY; [6]Human Brain Collection Core, Natl. Inst. of Mental Health, Bethesda, Bathesda, MD

Abstract: Dopaminergic neurons play a critical role for the neurobiology of disease, and treatment, in individuals with schizophrenia (SCZ) and Bipolar Disorder (BD). However, very little is known about genomic and transcriptomic alterations in midbrain dopaminergic neurons (mDN) including those residing in the Substantia nigra and Ventral tegmental area. To address this knowledge gap, we have developed a resource comprised of 111 RNA-seq and 94 HiC libraries, derived from FACS-sorted Nurr1+/NeuN+ immuno-tagged midbrain dopaminergic neuron, and their surrounding glial (Nurr1-/NeuN-) nuclei, from 36 SCZ, 19 BD and 56 control subjects. We identified 340 genes with dysregulated expression in mDN, while no significant gene changes were observed in non-neuronal cells from the same brain region. mDN dysregulated genes showed significant enrichment for GWAS-defined risk variants associated with SCZ, BD, and other psychiatric symptoms. Functional pathway analysis showed significant enrichment for genes regulating synaptic plasticity and ionl channel. Additionally, we conducted a distinct analysis utilizing one of the software tools developed in our laboratory to identify genes with transcripts that displayed discordant effect sizes. To better understand the regulation of these genes, we are currently exploring our mDN-specific Hi-C 3D genome maps to identify disease-relevant chromatin loops at the site of differentially expressed genes.. This approach provides further insights into the complex molecular landscape of SCZ. Overall, our research emphasizes the importance of dopamine neurons in midbrain and their dysregulation in SCZ, highlighting specific genetic and functional pathways implicated in the disorder.

Disclosures: **S. Singh:** None. **M. Iskhakova:** None. **T. Lambert:** None. **S. Marenco:** None. **P. Auluck:** None. **G. Hoffman:** None. **P. Roussos:** None. **K. Girdhar:** None. **S. Akbarian:** None.

Poster

PSTR177. Neuropathology, Genetics, Genomics, and Molecular Mechanisms: Schizophrenia

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR177.07/UU14

Topic: H.13. Schizophrenia

Title: Src tyrosine kinase deficient mice exhibit selective deficits in trace fear conditioning

Authors: *K. PAREKH[1], R. E. FEATHERSTONE[1], L. M. CROWN[1], C.-G. HAHN[2], M. W. SALTER[3], S. J. SIEGEL[1];
[1]Dept. of Psychiatry and Behavioral Sci., USC, Los Angeles, CA; [2]Dept. of Psychiatry & Neurosci., Thomas Jefferson Univ., Philadelphia, PA; [3]Neurosciences and Mental Hlth., Hosp. for Sick Children, Toronto, ON, Canada

Abstract: A growing body of evidence suggests that N-methyl-D-aspartate receptor (NMDAR) hypofunction is an important etiology in psychiatric disorders such as schizophrenia. The molecular underpinnings that lead to NMDAR hypofunction are not completely understood. Sarcoma tyrosine kinase (Src) is an important regulator of NMDAR activation via its influence on multiple schizophrenia susceptibility pathways (i.e., Dysbindin, neuregulin-1). Previous work done by our lab had shown that Src (+/-) heterozygous mice, which show reduced Src expression, display impaired working memory (WM) on a trace fear conditioning task (TFC). Moreover, impairments in TFC were alleviated following selective activation of synaptic Src via chronic administration of a TAT associated peptide. To further understand how Src regulates WM, we assessed c-Fos activation in wild-type (WT) and Src (+/-) mice following TFC. As a control, mice were also assessed on standard cued fear conditioning (sCFC). We assessed c-Fos both immediately following training as well as 48 hours after testing. Subjects included Src (+/-) and WT littermates tested on sCFC (WT: n=4; Src (+/-): n=4) and TFC (WT: n=7; Src (+/-): n=5). TFC was carried out in conventional chambers (Med Associates). TFC and sCFC procedures were identical with the exception that in sCFC, the unconditioned stimuli (US) terminated at the same time as the conditioned stimuli (CS) while in TFC there was a 20 second trace period separating CS from US. TFC was quantified as the ratio of time spent freezing during the cue versus pre-cue. We assessed c-Fos expression via immunohistochemistry in the hippocampus, mPFC, and amygdala. Relative to WT, Src (+/-) mice showed significantly reduced freezing during the CS 48 hours following TFC training ($p=0.004$). In contrast, Src (+/-) mice showed intact learning on sCFC ($p=0.7$). This pattern of results suggests a selective deficit in WM. We hypothesized that c-Fos expression will be reduced in the hippocampus and PFC in Src (+/-) mice during TFC, but not during sCFC. These results reinforce the importance of Src in regulating WM and support an important role for Src in cognitive impairment associated with mental illness. Future studies will assess the molecular mechanisms by which Src contributes to NMDAR related cognition.

Disclosures: K. Parekh: None. **R.E. Featherstone:** None. **L.M. Crown:** None. **C. Hahn:** None. **M.W. Salter:** None. **S.J. Siegel:** None.

Poster

PSTR177. Neuropathology, Genetics, Genomics, and Molecular Mechanisms: Schizophrenia

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR177.08/UU15

Topic: H.13. Schizophrenia

Title: Transcriptomic investigation in CRISPR/Cas9-mediated kainate receptor gene-knockout human neuroblastoma cells

Authors: *M.-C. CHENG;
Dept. of Psychiatry, Taipei Veterans Gen. Hosp. Yuli Br., Hualient Country, Taiwan

Abstract: The glutamate ionotropic kainate receptors, encoded by the GRIK gene family, are composed of four subunits and function as ligand-activated ion channels. They play a critical role in regulating synaptic transmission and various synaptic receptors' functions as well as in the pathophysiology of schizophrenia. However, their functions and mechanisms of action are poorly understood and worthy of exploring deep. To further understand the exact role of the kainate receptors *in vitro*, we generated kainate receptor-knockout (KO) SH-SY5Y cell lines by CRISPR/Cas9 mediated gene editing method and conducted RNA sequencing (RNA-seq) to determine the differentially expressed genes in the isogenic edited cells. To assess the morphology of edited cells, we induced edited SH-SY5Y cells into the differentiated cell types by sequentially treating retinoic acid and brain-derived neurotrophic factor and quantitated filamentous actin (F-actin) in edited cells using rhodamine-phalloidin staining. The RNA-seq and the Gene Ontology enrichment analysis revealed that genetic deletion of the *GRIK1*, *GRIK2*, and *GRIK4* genes disturbed multiple genes involved in multiple signal pathways, including a converging pathway related to the synaptic membrane. In the morphology study, the phase-contrast and fluorescent images demonstrated that less F-actin was expressed in differentiated SH-SY5Y cells harboring *GRIK1*, *GRIK2*, or *GRIK4* deficiency compared to wild-type cells. Our data indicate that kainate receptor deficiency can cause differential expression of genes involved in the synaptic membrane, and the elucidation of these genes should shed some light on the pathogenesis of schizophrenia. Furthermore, the transcriptomic profiles for kainate receptor-KO SH-SY5Y cells contribute to emerging evidence for the novel mechanisms underlying the effect of kainate receptors and molecular pathways of the pathophysiology of schizophrenia. Besides, our data suggest that kainate receptor-mediated F-actin remodeling may be a candidate mechanism underlying schizophrenia.

Disclosures: M. Cheng: None.

Poster

PSTR177. Neuropathology, Genetics, Genomics, and Molecular Mechanisms: Schizophrenia

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR177.09/UU16

Topic: H.13. Schizophrenia

Support: NIH Grant I01-CX001380-03

Title: Exploring segregating rare variants in familial whole genome sequencing data to elucidate genes associated with schizophrenia

Authors: *H. KAZEMI[1], J. DRAKE[2], T. BIGDELI[3], S. BACANU[4], K. BENKE[5], B. MAHER[5], C. CARVALHO[6], H. MEDEIROS[7], R. FERREIRA[8], J. KNOWLES[9], S. MCCARROLL[10], M. PATO[9], C. PATO[9], V. VLADIMIROV[1], A. FANOUS[1];
[1]Univ. of Arizona, Phoenix, AZ; [2]Texas A&M Univ., College Station, TX; [3]SUNY Downstate Med. Ctr., Brooklyn, NY; [4]Virginia Inst. for Psychiatric and Behavioral Genet., Richmond, VA; [5]Johns Hopkins Bloomberg Sch. of Publ. Hlth., Baltimore, MD; [6]Univ. of the Azores, Ponta Delgada, Portugal; [7]Keck Sch. of Med. of the Univ. of Southern California, Los Angeles, CA; [8]Inst. S. Joao de Deus, Lisbon, Portugal; [9]Rutgers Univ., Piscataway, NJ; [10]Harvard Med. Sch., Boston, MA

Abstract: Background: While genome-wide association studies (GWAS) have been successful in identifying common risk alleles contributing to schizophrenia etiology (SCZ), a highly heritable psychiatric disorder with substantially increased mortality risk, these studies often ignore the contribution of rare variants. Using whole genome sequencing (WGS) data from the Portuguese Island Collection (PIC) sample (n=175), which contains families with affected and unaffected individuals and controls, we performed two analytical approaches to assess the impact of common and rare variants. First, common variants were used to calculate the polygenic risk score (PRS) for SCZ. Secondly, a gene-based segregation test (GESE) was used to identify rare variants segregating in these families. **Methods**: Prior to analysis, summary statistics of gnomAD v3.1.2 were used to select 5,217,104 common single nucleotide polymorphisms (SNPs) with a MAF $\geq$0.01 and 5,706,926 rare single nucleotide variants (SNVs) with a MAF < 0.01. The selected rare variants localized to 17,346 protein-coding genes, and 411,434 appeared novel. The PRS was calculated using PRSice-2, using common variants in 58 SCZ patients and 62 control subjects. Only rare variants in families (n=20) containing affected (n=40) and unaffected (n=55) individuals were selected for GESE analysis. Lastly, functional annotation of segregating rare variants was conducted with ANNOVAR. **Results**: The p-value threshold of 0.05 explained the largest variation of risk for SCZ with $R^2 = 0.251$ (p-value = $2.2x10^{-5}$). GESE with ANNOVAR identified segregating variants leading to missense mutations in 3,940 genes, stop/gain mutations in 166 genes, and stop/loss mutations in 6 genes. Utilizing a weighted tabulation of each variant within the gene and the number of segregating families, the top genes were *RBFOX1*, *CTNNA3*, and *CSMD1*, all of which have been implicated in numerous psychiatric conditions, including SCZ. Additionally, these genes had variants segregated in multiple families, such as chr10:66742412 in *CTNNA3*, which was found in 6 families. **Conclusions**: By leveraging familial WGS, we were able to identify genes associated with SCZ. These results are preliminary and ultimately will need to be replicated in additional family-based samples.

Disclosures: H. Kazemi: None. **J. Drake:** None. **T. Bigdeli:** None. **S. Bacanu:** None. **K. Benke:** None. **B. Maher:** None. **C. Carvalho:** None. **H. Medeiros:** None. **R. Ferreira:** None. **J. Knowles:** None. **S. McCarroll:** None. **M. Pato:** None. **C. Pato:** None. **V. Vladimirov:** None. **A. Fanous:** None.

Poster

PSTR177. Neuropathology, Genetics, Genomics, and Molecular Mechanisms: Schizophrenia

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR177.10/UU17

Topic: H.13. Schizophrenia

Support: NIMH Grant 1R01MH125899

Title: Alterations in circHomer1 and Homer1 mRNA expression are associated with SNP risk alleles for schizophrenia and bipolar disorder

Authors: *M. KOCH, G. PAPAGEORGIOU, M. HERNANDEZ, G. MAXSON, M. OTERO, M. VARANGIS, N. MELLIOS;
Univ. of New Mexico Dept. of Neurosciences, Albuquerque, NM

Abstract: Schizophrenia (SCZ) and bipolar disorder (BD) are severe psychiatric disorders affecting approximately 1% and 3% of people in the United States, respectively. *Homer1* encodes scaffolding proteins involved in synaptic plasticity and has been implicated in the pathology of both disorders. Notably, *circHomer1*, the circularized isoform of the *Homer1* transcript and an activity-dependent regulator of *Homer1b*, is robustly reduced in post-mortem brains of patients with SCZ and BD. Recent genome-wide association studies (GWAS) of SCZ and BD have found associated single nucleotide polymorphisms (SNPs) in a putative enhancer region of *Homer1*. Here we find two risk alleles that are correlated with altered *circHomer1* and *Homer1* mRNA expression in the orbital frontal cortex (OFC) of patients with BD and SCZ, suggesting that these SNPs may play a functional role in regulation of *Homer1*. Future work will investigate the effect of these alleles on enhancer function and the contribution of this enhancer to *Homer1* gene expression. Together, this work will provide novel insights into genetic mechanisms that may contribute to altered *circHomer1* expression in patients with SCZ and BD.

Disclosures: M. Koch: None. **G. Papageorgiou:** None. **M. Hernandez:** None. **G. Maxson:** None. **M. Otero:** None. **M. Varangis:** None. **N. Mellios:** E. Ownership Interest (stock, stock options, royalty, receipt of intellectual property rights/patent holder, excluding diversified mutual funds); Circular Genomics Inc.

Poster

PSTR177. Neuropathology, Genetics, Genomics, and Molecular Mechanisms: Schizophrenia

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR177.11

Topic: H.13. Schizophrenia

Support: NSFC Grant 82090031

Title: Unravel the molecular and cellular signatures of schizophrenia by single-cell multi-omics

Authors: *X. CHEN, B. YU, Y.-H. XU, X.-M. LI;
Zhejiang Univ., Hangzhou, China

Abstract: Schizophrenia is a complex and severe psychiatric disorder that has a profound effect on both the individuals affected and society. Despite decades of research, our understanding of the molecular and cellular mechanisms underlying schizophrenia remains limited. To unravel the vulnerability in schizophrenia at the single-cell level, we performed snRNA-seq and snATAC-seq on nuclei isolated from human dorsolateral prefrontal cortex (dlPFC) of both males and females. The fresh frozen postmortem brain tissues were from patients with schizophrenia as well as cognitively healthy matched controls. Totally, 27,188 nuclei for snRNA-seq and 19,512 nuclei for snATAC-seq were collected. We identified 22 cell types with specific gene expression patterns and chromatin landscapes in order to construct a single-cell atlas of schizophrenia-altered genes and regulatory dynamics. In addition, spatially resolved transcriptomics (Stereo-seq) further verified upper-layer (especially Layer 2-3 IT) neuron vulnerability in schizophrenia. Lastly, We mapped rare and common psychiatric disorder-associated variants to specific cell types and cis-regulatory elements. Overall, our high-resolution multi-omic single cell atlas provides valuable resources for understanding the molecular and cellular alterations associated with schizophrenia, links cell type-specific transcriptomic and epigenetic changes to etiological genetic risk factors, which could ultimately lead to the development of new therapeutic approaches.

Disclosures: **X. Chen:** None. **B. Yu:** None. **Y. Xu:** None. **X. Li:** None.

Poster

PSTR177. Neuropathology, Genetics, Genomics, and Molecular Mechanisms: Schizophrenia

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR177.12/Web Only

Topic: H.13. Schizophrenia

Support: NIH Grant MH109260
NIH Grant T32 MH014654
Pennsylvania Department of Health Tobacco Settlement Act Grant RFA67-76
2017 NARSAD Young Investigator Grant #26634

Title: Differential gene expression and transcript usage in a mouse model promotes characterization of schizophrenia pathophysiology

Authors: *A. E. WELLER[1], T. N. FERRARO[2], G. A. DOYLE[3], B. C. REINER[1], W. H. BERRETTINI[1], R. C. CRIST[1];
[1]Univ. of Pennsylvania, Philadelphia, PA; [2]Cooper Med. Sch. of Rowan Univ., Camden, NJ; [3]Fox Chase Cancer Center, Temple Univ. Hlth. Syst., Philadelphia, PA

Abstract: *Setd1a* is a gene implicated in schizophrenia, thus the study of heterozygous *Setd1a*$^{+/-}$ knockout mice may provide useful insight into disease pathogenesis. Employing a publicly available single-cell RNA sequencing (scRNAseq) dataset, we used Seurat and Sierra analytic R packages to identify differentially expressed genes (DEGs) and differential transcript usage (DTU), respectively, in individual cell types from the prefrontal cortex (PFC) and striatum of *Setd1a*$^{+/-}$ mice compared to wild-type controls. We further analyzed cell type-specific DEGs and genes containing DTU using Ingenuity Pathway Analysis (IPA). We identified 377 unique DEGs across all cell types in PFC and 677 unique gene peaks containing DTU. In striatum, we identified 327 unique DEGs across all cell types and 9 unique gene peaks containing DTU. IPA analysis of DEGs resulted in identification of 15 canonical pathway terms that appeared in more than one cluster in PFC; 25 canonical pathway terms appeared in more than one cluster in striatum. IPA analysis of genes containing DTU resulted in 29 canonical pathway terms that appeared in more than one cluster in the PFC dataset. No canonical pathways terms appeared in striatum. IPA findings from our separate DEG lists in PFC and striatum highlight the importance of cell metabolism, immune response/inflammation, mitochondrial function, and protein translation in schizophrenia. IPA analysis of genes containing DTU in PFC points to alteration in cellular processes including intracellular signaling, neurotransmission, and protein synthesis/degradation. One canonical pathway, 'EIF2 Signaling,' which is involved in the regulation of protein synthesis, was detected in PFC DEGs, striatum DEGs, and PFC genes containing DTU, drawing attention to its importance in schizophrenia pathophysiology. The 'EIF2 Signaling' pathway could be a target for the development of new treatments and biomarkers for schizophrenia, but additional studies are needed to determine the specific role of 'EIF2 Signaling' in schizophrenia etiology.

Disclosures: A.E. Weller: None. **T.N. Ferraro:** None. **G.A. Doyle:** None. **B.C. Reiner:** B. Contracted Research/Research Grant (principal investigator for a drug study, collaborator or consultant and pending and current grants). If you are a PI for a drug study, report that research relationship even if those funds come to an institution.; Research support from Novo Nordisk and Boehringer Ingelheim that was not used in support of this study. **W.H. Berrettini:** None. **R.C. Crist:** None.

Poster

PSTR177. Neuropathology, Genetics, Genomics, and Molecular Mechanisms: Schizophrenia

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR177.13/UU18

Topic: H.13. Schizophrenia

Support: NIH Grant 1R21MH126328-01

Title: Mitochondrial function and gene expression as a predictive biomarker of schizophrenia-related symptoms in 22q11.2 deletion syndrome

Authors: D. FREDERICK[1], T. CROWLEY[2], S. GALLAGHER[2], O. TRAN[1], ***M. KIM**[1], B. EMANUEL[1], D. MCDONNALD-MCGINN[1], D. WALLACE[1,3], R. GUR[2], C. GIULIVI[3], S. ANDERSON[2];
[1]Children's Hosp. of Philadelphia, Philadelphia, PA; [2]Univ. of Pennsylvania, Philadelphia, PA; [3]Univ. of California, Davis, Davis, CA

Abstract: The 22q11.2 deletion syndrome (22q11DS) is associated with a roughly 25% risk of developing schizophrenia (SZ)-related symptoms. Since the features of SZ in the context of 22q11DS are shared with non-syndromic SZ, the high rate of SZ in 22q11DS provides an opportunity for longitudinal studies that identify cognitive and physiological changes that predict SZ risk. Such identification can lead to mechanistic studies behind the variable penetrance (VP) for SZ in 22q11DS, and lead to preventative measures. Multiple lines of evidence from studies of human blood, human genetics, IPSC-derived neurons (iNs), and mouse models, suggest involvement of mitochondrial (mito) dysfunction. Indeed, 6 of the 46 genes in the deleted region encode for mito localizing proteins. Using iNs, we found that while OXPHOS is reduced in the 22q11DS with SZ (22q(+)SZ) group relative to control (CTRL)(Li et al., 2019), the 22q11DS without SZ (22q(-)SZ) group has CTRL-levels of OXPHOS (Li et al., 2021). Relative to both CTRL and 22q(+)SZ groups, the 22q(-)SZ group has upregulated expression of PGC1α, a "master regulator" of mito biogenesis, and multiple genes involved in OXPHOS (Li et al., 2021). These results suggest that VP for SZ in 22q11DS may be influenced by an individual's capacity for mito compensation. To test this idea with a higher throughput approach than possible with iNs, we examined 20 lymphoblastoid cell lines (LCLs) from 22q11DS adults, equally split between 22q(+)SZ and 22q(-)SZ. By analysis of a few measures of OXPHOS activity and related gene expression, we found that mito complex I activity (COI) is higher in the 22q(-)SZ group, as are the expression levels of the COI gene NDUFV1 and PGC1α along with its co-factor PPARa (Li et al, 2021). These results raise the question, can failed mito compensation identified in LCLs from teenagers with 22q11DS predict their likelihood of developing SZ in later adolescence or early adulthood? In this study, we are examining mito function in LCLs from individuals with 22q11DS, without SZ-related symptoms at the time LCLs were generated, and for whom we have follow-up clinical assessments into their 20s. Proteomic and oximetry assessments suggest, based on N=5 “converters” to SZ and 5 “non-converters,” that the non-converter group has better mito energetics than the converter group. Studies are ongoing to increase the number of subjects. The identification of a metabolic, blood-based biomarker of SZ “super-risk” in 22q11DS would be invaluable, particularly since there are FDA-approved medications that enhance mito biogenesis, and thus might be used to prevent the development of SZ in 22q11.2 deletion syndrome.

Disclosures: D. Frederick: None. **T. Crowley:** None. **S. Gallagher:** None. **O. Tran:** None. **M. Kim:** None. **B. Emanuel:** None. **D. Mcdonnald-Mcginn:** None. **D. Wallace:** None. **R. Gur:** None. **C. Giulivi:** None. **S. Anderson:** None.

Poster

PSTR177. Neuropathology, Genetics, Genomics, and Molecular Mechanisms: Schizophrenia

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR177.14/UU19

Topic: H.13. Schizophrenia

Support: Lieber Institute for Brain Development

Title: Investigating the impact of common genetic risk variants for schizophrenia on neuronal function elucidates relationships between cellular phenotypes, clinical status and cognitive performance

Authors: *G. SHIM[1], S. C. PAGE[1], S. R. SRIPATHY[1], F. FARINELLI[1], Z. YE[1], Y. WANG[1], D. DAS[1], D. J. HILER[1], E. A. PATTIE[1], C. V. NGUYEN[1], M. TIPPANI[1], H.-Y. CHEN[1], M. N. TRAN[1,2], N. J. EAGLES[1], J. M. STOLZ[1], J. L. CATALLINI[1,3], O. R. SOUDRY[1], D. DICKINSON[5], D. R. WEINBERGER[1,2,6,7,8], K. MARTINOWICH[1,6,7], M. L. MACDONALD[9], J. SHIN[1], A. E. JAFFE[1,3,6,7,4], R. E. STRAUB[1], B. J. MAHER[1,6,7];
[1]Lieber Inst. for Brain Develop., Baltimore, MD; [2]Dept. of Genet. Med., McKusick-Nathans Inst., Baltimore, MD; [3]Dept. of Biostatistics, [4]Dept. of Mental health, Johns Hopkins Bloomberg Sch. of Publ. Hlth., Baltimore, MD; [5]Clin. and Translational Neurosci. Br., NIMH/NIH, Bethesda, MD; [6]The Solomon H Snyder Dept. of Neurosci., [7]Dept. of Psychiatry and Behavioral Sci., [8]Dept. of Neurol., Johns Hopkins Sch. of Med., Baltimore, MD; [9]Dept. of Psychiatry, Univ. of Pittsburgh, Pittsburgh, PA

Abstract: Schizophrenia (SCZ) is a complex, polygenic disorder with marked clinical heterogeneity and no clear pathological mechanism or cellular pathology. Previous human induced pluripotent stem cell (hiPSC) studies have identified cellular phenotypes associated with SCZ diagnosis, yet their relevance to the clinical and cognitive features remains uncertain. Here, we describe results of our investigation into the relationships between cellular measures obtained from hiPSC-derived neurons and the individual's clinical status, severity of symptoms, and cognitive performance. We further identify within-case patterns of association that could serve to illuminate dimensions of illness and subgroups of patients that may be suitable for targeted treatments. In hiPSC-derived neurons from 13 high polygenic risk score (PRS) SCZ patients and 15 low PRS neurotypical controls (CON), we observed several electrophysiological measures related to Na^+ channel function that were associated with diagnosis of SCZ. Lines derived from SCZ donors showed an increased membrane resistance, increased number of Na^+ current peaks in response to a voltage ramp protocol, shifted activation threshold of the second Na^+ peak, significantly hyperpolarized voltage dependence of activation and inactivation, reduced long term inactivation, and decreased action potential interspike interval. In SCZ, the number of Na^+ peaks showed a positive association with the severity of hallucinations, and the sEPSC amplitude showed a negative correlation with performance on the Wisconsin Card Sort Test. Additionally, we are generating human forebrain organoids (FBOs) from these hiPSC lines and performing extensive cellular characterization using immunofluorescence staining, transcriptomic profiling, proteomics, and electrophysiology to find novel association with clinical phenotypes. Remarkably, day 150 SCZ FOBs showed similar hyperpolarized shifts in the voltage dependence of activation and inactivation. These consistent findings across two distinct differentiation protocols suggest that the kinetic properties of Na^+ channels may represent a convergent

biophysical mechanism downstream of common variant risk for SCZ and underscore the potential of this approach for biomarker identification and downstream drug development.

Disclosures: **G. Shim:** None. **S.C. Page:** None. **S.R. Sripathy:** None. **F. Farinelli:** None. **Z. Ye:** None. **Y. Wang:** None. **D. Das:** None. **D.J. Hiler:** None. **E.A. Pattie:** None. **C.V. Nguyen:** None. **M. Tippani:** None. **H. Chen:** None. **M.N. Tran:** None. **N.J. Eagles:** None. **J.M. Stolz:** None. **J.L. Catallini:** None. **O.R. Soudry:** None. **D. Dickinson:** None. **D.R. Weinberger:** None. **K. Martinowich:** None. **M.L. MacDonald:** None. **J. Shin:** None. **A.E. Jaffe:** None. **R.E. Straub:** None. **B.J. Maher:** None.

Poster

PSTR177. Neuropathology, Genetics, Genomics, and Molecular Mechanisms: Schizophrenia

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR177.15/UU20

Topic: H.13. Schizophrenia

Support: NIMH Intramural Research Program
NIH Grant K22 MH126015

Title: An atlas of cell types in the human mediodorsal thalamus

Authors: ***A. SCHULMANN**[1], S. MARENCO[1], P. AULUCK[1], N. FENG[1], Q. XU[1], Y. PATEL[1], A. MUKHERJEE[2], R. KOMAL[1], Y. LENG[1], C. GAO[1], N. AKULA[1], G. DUGAN[1], S. WILLIAMS[1], R. EDELMANN[1], S. ROY[1], T. USDIN[1], J. SINGH[3], M. KELLY[3], M. HALASSA[2], S. HATTAR[1], M. PENZO[1], F. MCMAHON[1];
[1]NIMH, Bethesda, MD; [2]Tufts Univ., Boston, MA; [3]NCI, Bethesda, MD

Abstract: The mediodorsal thalamus (MDT) and its connections to the prefrontal cortex (PFC) are crucial for many executive functions such as working memory and cognitive control. MDT-PFC connectivity is reduced consistently and early on in individuals with psychotic spectrum illness. To understand the neurobiological basis of these changes, it is important to characterize the cell types in the human MDT. Here we investigated the cell type composition of human MDT in 10 neurotypical individuals from two brain banks. For comparison, we also analyzed mouse MDT pooled from 4 male and 4 female mice. We isolated nuclei from these samples and performed single-nucleus RNA sequencing on the 10X Chromium platform. Data was processed via *CellRanger*, filtered, and clustered in *Seurat*. For spatial mapping, we used RNAscope HiPlex fluorescent *in situ* hybridization (RNA-FISH) on coronal thalamic sections, followed by imaging and removal of lipofuscin signal. Additionally, we used spatial transcriptomics on thalamic sections using 10X Visium and analyzed the data using *SpaceRanger* and *Seurat*. Excitatory neurons in human MDT exhibited a gradient associated with genes related to synaptic signaling and ion transport, analogous to a gradient previously found across mouse thalamus, with narrow-projecting cells on one end (*primary*) and broad-projecting cells on the other end

(*secondary*/*tertiary*). Inhibitory neurons in human MDT comprised over a third of all thalamic neurons and expressed *SOX14* and *OTX2*, consistent with midbrain-derived developmental origin. Integration of mouse and human MDT identified similar cell types between the species. Notable differences included the absence of inhibitory neurons in mouse MDT and a larger proportion of *primary* neurons in human MDT. Spatial transcriptomics and RNA-FISH showed that *primary* thalamic markers localized within the MDT body, while those of *tertiary* thalamic cells mapped to the midline and along the lateral border of MDT. Our study highlights the presence of diverse cell populations in human MDT and provides a map of cell types for future case control studies. Excitatory neurons exhibit a major gradient, which parallels a known thalamus-wide functional gradient. Cross-species analyses suggest that *primary* neurons are expanded in human MDT relative to mice. Spatial mapping showed segregation of excitatory populations with *primary* neurons located within the MDT body and *tertiary* neurons in the midline/paraventricular thalamus and intralaminar nuclei. Future work will explore links between thalamic gene expression and psychiatric disease risk through integration with genome-wide association studies of serious mental illness.

Disclosures: **A. Schulmann:** None. **S. Marenco:** None. **P. Auluck:** None. **N. Feng:** None. **Q. Xu:** None. **Y. Patel:** None. **A. Mukherjee:** None. **R. Komal:** None. **Y. Leng:** None. **C. Gao:** None. **N. Akula:** None. **G. Dugan:** None. **S. Williams:** None. **R. Edelmann:** None. **S. Roy:** None. **T. Usdin:** None. **J. Singh:** None. **M. Kelly:** None. **M. Halassa:** None. **S. Hattar:** None. **M. Penzo:** None. **F. McMahon:** None.

Poster

PSTR177. Neuropathology, Genetics, Genomics, and Molecular Mechanisms: Schizophrenia

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR177.16/UU21

Topic: H.13. Schizophrenia

Support: R01MH110921
R01MH075916
P50MH096891

Title: Postmortem proteomic analysis of glutamatergic receptor complexes in schizophrenia

Authors: *A. MARC[1,2], H. LIN[1,2], M. MSACKYI[1,2], W. ZHANG[1,2], K. BORGMANN-WINTER[1,2,3], C.-G. HAHN[1,2,3];
[1]Neurosci., Farber Inst. of Neurosci. Thomas Jefferson Univ., Philadelphia, PA; [2]Dept. of Neurosci. Thomas Jefferson Univ., Philadelphia, PA; [3]Dept. of Psychiatry and Behavioral Sci. Thomas Jefferson Univ., Philadelphia, PA

Abstract: The glutamatergic synapse has long been identified as a locus where multitudes of altered molecules and pathways intersect to precipitate the pathology of schizophrenia (SCZ).

Indeed, the emerging genetic architecture of SCZ reveals a striking enrichment of risk genes in the synapse, particularly in the postsynaptic density (PSD) where glutamatergic receptors reside. Increasing evidence supports that physical interactions between proteins (protein interactions) are mechanistic substrates by which risk gene products interact and converge in the spine synapse in SCZ. To date, most postmortem studies of glutamatergic signaling have focused on the levels of mRNA or protein expression, but not protein interactions. Our goal is to identify altered protein interactions by delineating the protein composition of glutamatergic receptor complexes in postmortem brains of SCZ using immunoprecipitation-mass spectrometry (IP-MS). Synaptosomes were isolated from human post-mortem dorsolateral-prefrontal cortex (DLPFC) from healthy controls and age-sex matched patients. Synaptosomal extracts were immunoprecipitated (IP) for NMDA receptor subunit 1 (GluN1) or metabotropic glutamate receptor 5 (mGluR5), two receptors for which reciprocal interaction has previously been demonstrated. Western Blot (WB) analysis was conducted for IP bait proteins and known binding partners (PSD-95, PLCy1, NMDAR2A) prior to Trypsin/Lys-C digestion into peptides for Selected Reaction Monitoring Mass Spectrometry (SRM-MS). To identify proteins in the receptor complexes, we surveyed 2298 peptides representing 287 synaptic proteins using IP-SRM-MS. GluN1 complexes contained peptides representing 129 proteins while mGluR5 complexes consisted of 112 proteins among the surveyed. As a preliminary analysis, a subset of 5 patients and their age-sex-matched controls were examined for differentially represented proteins and their enrichment in pathways in patients compared to controls. GluN1 complexes derived from patients exhibited altered levels of 20 proteins ($p<0.1$) including GRIN2A, GRM5, CAMK2A, and CAMK2B, showing enrichment for calcium and glutamatergic signaling in pathway analyses. mGluR5 complexes exhibited altered levels of 17 proteins ($p<0.1$) including CAMK2A, GRIA2, GRIN1, GRIN2B, and GRM2, showing enrichment for the pathways of glutamatergic signaling and postsynaptic density. Thus, the IP-SRM-MS analysis of subcellular fractions enables us to delineate protein composition of postmortem glutamatergic receptor complexes and identify altered protein interactions as points of convergence in schizophrenia pathophysiology.

Disclosures: **A. Marc:** None. **H. Lin:** None. **M. Msackyi:** None. **W. Zhang:** None. **K. Borgmann-Winter:** None. **C. Hahn:** None.

Poster

PSTR177. Neuropathology, Genetics, Genomics, and Molecular Mechanisms: Schizophrenia

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR177.17/UU22

Topic: H.13. Schizophrenia

Support: MH132097
MH059852
NIAAA

Title: Direct Conversion of Somatic Cells to Retain Donors' Epigenetic Landscape: Olfactory Epithelial Cells vs Skin Fibroblasts

Authors: *M. MSACKYI[1,2], H. LIN[1], W. ZHANG[1,2], A. MARC[1,2], C.-G. HAHN[1,2,3], K. BORGMANN-WINTER[3,1,2];
[1]Neurosci., [2]Vickie & Jack Farber Inst. for Neurosci., Thomas Jefferson Univ., Philadelphia, PA; [3]Dept. of Psychiatry and Behavioral Sci. & Neurosci., Sidney Kimmel Med. Col. at Thomas Jefferson Univ., Philadelphia, PA

Abstract: Common neuropsychiatric illnesses, such as psychotic, mood or developmental disorders, have complex pathophysiologic mechanisms. These mechanisms involve multitudes of common and rare genetic variants modified by epigenetics to produce a vast array of mRNA transcripts and proteins leading to neuropsychiatric illness. Using in vitro models is effective for both drug discovery and the testing of currently used drugs to reveal the mechanisms of neuropsychiatric illnesses. For the past decade, iPSCs have led the field of in vitro modeling of neuropsychiatric illnesses. iPSC derived neurons recapitulate the ontogeny of neural cells, offering a powerful paradigm to decipher neurodevelopmental processes. However, iPSCs largely erase the epigenetic characteristics of donors associated with neuropsychiatric illnesses during the process of stem cell generation. However, this issue is bypassed using direct conversion of somatic cells into neurons (DCiNs). This method bypasses the stem cell stage maintaining more epigenetic characteristics of the source cells. The epigenetic landscape of DCiNs is determined by two key factors; a) source cell types, and b) conversion protocols. Currently skin fibroblasts (SFs) are used commonly for DCiNs. However, SFs may be unable to retain the epigenetics of neuropsychiatric illnesses because SFs are non-neural cells. Olfactory neuroepithelial cells (OEs) are the only readily obtainable neural cells from patients. OEs also retain several neurobiological characteristics of neuropsychiatric illnesses. Here we compare SFs and OEs as source cells for a DCiN paradigm using SFs and OEs taken from the same individuals. ATAC-sequencing was performed on undifferentiated SFs and OEs to assess the epigenome. Results revealed a vast difference in the open chromatin regions (OCRs) between the two cell types, with the OCR parameters much more enhanced in OEs than SFs. OEs specific open chromatin was in many key regions associated with neuropsychiatric illness. Both SFs and OEs were then differentiated into DCiNs (DCiN-SF and DCiN-OE respectively) and were compared for their differing rates of neuronal maturation. DCiN-OE express MAP2 and synapsin proteins at a higher level than DCiN-SF at the same day in vitro. Patch clamp electrophysiology has shown development of strong inward currents at an earlier date in DCiN-OE compared to DCiN-SF. Finally, single cell-RNA-sequencing has been conducted comparing DCiN-OE and DCiN-SF. Taken together, our results suggest that OEs may serve as source cells for direct neuronal conversion that are more likely to reflect the epigenetic characteristics of subjects with neuropsychiatric illnesses.

Disclosures: M. Msackyi: None. **H. Lin:** None. **W. Zhang:** None. **A. Marc:** None. **C. Hahn:** None. **K. Borgmann-Winter:** None.

Poster

PSTR177. Neuropathology, Genetics, Genomics, and Molecular Mechanisms: Schizophrenia

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR177.18/UU23

Topic: H.13. Schizophrenia

Support: RO1MH110921
RO1 MH075916
P50MH096891

Title: Mice treated with risperidone during the juvenile period have long term effects on social memory and PFC neuronal activity in adulthood

Authors: *C.-G. HAHN[1], W. ZHANG[3], M. MSACKYI[2], A. WU[3], K. BORGMANN-WINTER[3];
[1]Thomas Jefferson Univ., Bryn Mawr, PA; [2]Thomas Jefferson Univ., Philadelphia, PA; [3]Thomas Jefferson university, Philadelphia, PA

Abstract: Adolescence is a crucial stage for the development of exploratory behaviors and cognitive control functions. Over the last decade, prescription of antipsychotics for off-label indications in adolescence has drastically increased, affecting almost 2% of persons under 18. Moreover, a higher proportion of young male patients are administered antipsychotic medications for off-label use and for longer durations of time. Given the vulnerability of the developing brain, a pressing concern is off-target CNS effects of these agents which may be sustained beyond the treatment period. Neurobiological effects of antipsychotics have been extensively studied but mostlyin adulthood, but not in adolescence. During this highly dynamic period of brain development, we asked how antipsychotics affect exploratory and cognitive behaviors and brain circuits acutely and chronically. In this study, we treated WT mice with IP injection of risperidone(4mg/kg) (n=16) or saline (n=14) daily from P28-P49 (21 days). During adulthood (P62-P110), we tested the effect of risperidone on working memory, social memory, anxiety levels and spatial memory using Y-maze, three chamber social interaction, open field and novel object recognition. We found that the mice that were treated with risperidone during the juvenile period showed decreased social recognition and increased anxiety compared to those treated with the vehicle in the adulthood. Notably, male mice showed greaterimpairment compared to the female animals in social recognition test and open field test. We then examined these mice by employing In vivo Ca2+ imaging using two photon microscope focusing on layer 2 neurons ofthe M2 cortex of awake animals during adulthood. Mice treated with risperidone during the juvenile period demonstrated decreased frequency in neuron firing compared to controls. Taken together, our results suggest that adolescent risperidone treatment may induce long-term changes in social memory and anxiety levels, associated with altered neuronal activity in the frontal cortex. Our results, while preliminary, may offer additional consideration in off-label treatment of adolescents with antipsychotics and for the possible relevance of sex differences in these effects.

Disclosures: C. Hahn: None. **W. Zhang:** None. **M. Msackyi:** None. **A. Wu:** None. **K. Borgmann-Winter:** None.

Poster

PSTR177. Neuropathology, Genetics, Genomics, and Molecular Mechanisms: Schizophrenia

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR177.19/UU24

Topic: H.13. Schizophrenia

Title: A REVIEW: INVESTIGATION OF THE ROLE OF SECOND-GENERATION ANTIPSYCHOTICS IN THE FORMATION OF METABOLIC SYNDROME AND SYSTEMATIC LITERATURE REVIEW OF THE RELATIONSHIP BETWEEN SEROTONERGIC GENE TYPES.

Authors: *N. ÇAVUS;
Uskudar Univ., ISTANBUL, Turkey

Abstract: ABSTRACTA Review: Investigation of the role of second generation antipsychotics in the formationof metabolic syndrome and systematic literature review of the relationship betweenserotonergic gene types.Objective: It was found out that serotonin receptors of serotonin receptor antagonist drugsused in schizophrenia are also present in the cardiovascular system. The aim of this study is toexamine the relationship between the drug active substances, which are serotonin antagonists,and the rate of cardiovascular diseases in patients with schizophrenia.**Methods:** The aim of this used study is to contribute to the development of the best practicebecause of the pharmacogenetic properties of the drugs in the treatment by combining bothgenetic and structural causes of a complex syndrome that shows the symptoms of manydiseases such as schizophrenia.Cohort, case-control, double-blind, placebo control, semi-experimental, controlled clinicalstudies, drug efficacy studies in patients with schizophrenia have been seen in cardiovasculardiseases because of the interaction of 5 HT genes and polymorphisms with the atypicalantipsychotics' risperidone, olanzapine, and aripiprazole active ingredients. A meta-analysisstudy will be conducted by calculating the obesity-related mortality rate with SPSS and lookingat the correlation between them. Before starting the meta-analysis, we will evaluate whetherthe publication bias does not affect the results of the study. To identify the studies to be selectedlater, search engines are ISI Web of Science, Cochrane Library, Web of Knowledge, NationalInstitute for Health and Clinical Evidence (NICE), Pubmed, PsychInfo, Scopus, EBSCO Host,Blackwell Publishing, Embase, Google Academic, Genetic. We will use the data searchengines Findings GWAS, copy number of Variations (CNVS), Association and LinkageStudies, Encyclopedia of DNA Elements (ENCODE). All studies, studies and data carried outbetween 1990 and January 2022 will be scanned, studies based on quantitative observationswill be selected among the cohort studies conducted during this period and a coding file willbe created after collecting the studies, pilot coding is carried out and the data are calculatedaccording to the coding file, and the main analyzes are done. In addition to the search engines, we will use in the literature review, theses, compilationarticles, bibliographies, manual searches in related journals, reports and bibliographies werescanned. The data, which we obtained using Boolean and fuzzy logic methods, are onlyquantitative studies and those whose

publication language is English and between 1990 and2022 were selected.**Keywords:** We chose to conduct this study are Drug Interactions, HTR2A, MetabolicSyndrome, Schizophrenia were figured out as.

Disclosures: N. Çavus: None.

Poster

PSTR177. Neuropathology, Genetics, Genomics, and Molecular Mechanisms: Schizophrenia

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR177.20/UU25

Topic: G.08. Other Psychiatric Disorders

Support: CIHR grant PJT-173287

Title: Characterizing von economo neurons in schizophrenia and major depressive disorder

Authors: *C. CANONNE[1,2], A. AGHABY-CLOUTIER[2], F. MECHAWAR[2], M.-A. DAVOLI[2], L. PALANYIAPPAN[2,3], N. MECHAWAR[2,3];
[1]Psychiatry, McGill Univ. Integrated Program in Neurosci., Montreal, QC, Canada; [2]Douglas institute, Montreal, QC, Canada; [3]Psychiatry, McGill Univ., Montreal, QC, Canada

Abstract: Background : Von Economo neurons (VENs) are a type of bipolar spindle-shaped neurons found in some regions of the human brain, including the anterior cingulate cortex (ACC; layer Vb) and frontal insula (FI). These neurons, absent in rodents, have been described in a few other species with large brains and complex social structures, and proposed to be involved in the regulation of negative emotions. An alteration in the number and morphology of VENs has been reported in several brain disorders including schizophrenia (SCZ) and suicide in psychosis. Recently, a transcriptomic analysis of VENs enabled the identification of differentially expressed genes in VENs vs pyramidal neurons, in particular a higher expression of genes associated with psychiatric and neurological disorders, including SCZ and depression. However, the implication of VENs in neuropsychiatric disorders remains to be largely characterized. While SCZ and MDD are two severe mental disorders affecting a significant portion of the population, the underlying biological causes remain ill-defined, preventing the development of more efficient treatments. A better comprehension of VENs implication in those two mental illnesses could lead to better therapies. With this project, we aim to identify specific VEN features that may be associated with major depressive disorder (MDD) and SCZ. We hypothesize that VEN densities, morphologies, and transcriptomic profiles display illness-specific alterations. **Methods** : Well-characterized post-mortem brain samples from males and females having died with MDD or SCZ were provided by the Douglas-Bell Canada Brain Bank. The densities of VENs in the FI and ACC are manually analyzed at a 20x magnification using QuPath. The density of layer Vb cells is also be determined manually to evaluate whether any significant difference is due to VENs specifically or, more generally, to layer Vb cells. Soma diameters is manually calculated

using the annotation tool for each individual VEN. **Results** : Preliminary results on VEN densities in the ACC of a subset equally distributed with male and female of MDD samples (n=10) revealed no significant difference ($p > 0.05$) compared to matched controls (n=10) with respectively a mean of 359.30 and 257.20 number of VENs per mm^3 and a standard deviation of respectively 209.64 and 232.41 with a one-way ANOVA. Current quantifications for the whole sample as well as for the FI and the SCZ group will be presented. **Conclusion** : Together, these results should provide a better understanding of the cerebral changes that occur in MDD and SCZ and shed new light on the possible implication of VENs in mental illness.

Disclosures: C. Canonne: None. **A. Aghaby-Cloutier:** None. **F. Mechawar:** None. **M. Davoli:** None. **L. Palanyiappan:** None. **N. Mechawar:** None.

Poster

PSTR177. Neuropathology, Genetics, Genomics, and Molecular Mechanisms: Schizophrenia

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR177.21/UU26

Topic: G.08. Other Psychiatric Disorders

Support: Unrestricted Grant from HLS Therapeutics to Sylvain Grignon MD PhD

Title: A pilot study of brain neurosteroid and steroid levels in a murine, two hit model of suicide in schizophrenia

Authors: M. ARGUIN[1], L. HAROUNE[2], S. SAIBI[2], E. COLOMBO[2], M.-C. DURPES[1], E. MARSAULT[2], ***S. GRIGNON**[1];
[1]Psychiatry & Pharmacology/physiology, Univ. De Sherbrooke, Sherbrooke, QC, Canada; [2]Sherbrooke Pharmacol. Inst., Univ. de Sherbrooke, Sherbrooke, QC, Canada

Abstract: Suicide is one of the leading causes of premature mortality in schizophrenia, with distinct pathophysiological mechanisms compared to mood disorders. While lithium has to been shown to decrease suicidal behavior (SB) in bipolar disorder, the antipsychotic drug clozapine (CLZ) is especially effective in improving SB in schizophrenia. Using a two-hit model of suicide in schizophrenia (THMS), we previously showed that maternal gestational immune activation (MIA) followed by social isolation (SI) increased helplessness, impulsivity and aggression, which were normalized by CLZ but not lithium. Conversely, SI alone, generally used to model mood disorders, elicited SB that were responsive to lithium but relatively unchanged by CLZ, thereby mimicking the differential responses in clinical populations. Subsequently, we showed that some of the effects of CLZ could be prevented by the 5-alpha reductase inhibitor finasteride, suggesting the involvement of neurosteroids (NS) in the effects of CLZ. This prompted us to measure the levels of some NS in THMS and control mice (Ct). *Methods.* THMS (MIA + SI) (n=8) and Ct (sham injection, no SI) (n=6) were constituted as previously described (PMID **29630964**). NS assay : An LC-MS/MS methodology was developed and validated for the

analysis of 8 NS (allopregnanolone (ALLO), pregnenolone (PREG), pregnenolone-S, dehydroepiandrosterone (DHEA), DHEA-S, progesterone, 3α-androstanediol and androsterone)). Briefly, a 20 mg sample was homogenized and extracted with 200 µL of acetonitrile. After agitation, the organic layer was collected and evaporated to dryness. The extract was then reconstituted in 30 µL of H_2O: MeOH (1:1) and filtered through 0.22 µM PTFE syringe filter before analysis. *Results.* Among the NS assayed, ALLO and PREG could be reliably identified in whole brain extracts. Their levels did not differ significantly in brain or plasma between the THMS and Ct. Progesterone and cortisol were decreased, versus Ct, in THMS in brain (-54% and -30%, respectively, $p<0.001$) and plasma (-49% and -37%, respectively, $p<0.001$). *Discussion*. There were no significant differences in the levels of ALLO and PREG in the whole brain. These findings suggest that SB, and the effects of CLZ on SB, could rather involve regional differences. Further investigations are warranted to explore the regional distribution of NS and their implication in the context of suicide in schizophrenia.Interestingly, we observed a significant decrease in progesterone and corticosterone levels in the THMS compared to controls. This result could be attributed to an as yet unexplained interaction between MIA and SI.

Disclosures: **M. Arguin:** None. **L. Haroune:** None. **S. Saibi:** None. **E. Colombo:** None. **M. Durpes:** None. **E. Marsault:** None. **S. Grignon:** B. Contracted Research/Research Grant (principal investigator for a drug study, collaborator or consultant and pending and current grants). If you are a PI for a drug study, report that research relationship even if those funds come to an institution.; Unrestricted Grant from HLS Therapeutics Canada.

Poster

PSTR177. Neuropathology, Genetics, Genomics, and Molecular Mechanisms: Schizophrenia

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR177.22/UU27

Topic: G.08. Other Psychiatric Disorders

Support: 1K99MH 129617-01A1

Title: A novel circuit underlying amotivation in the 22q11DS model of schizophrenia

Authors: *M. H. PATTON, B. J. W. TEUBNER, S. S. ZAKHARENKO;
Developmental Neurobio., St. Jude Children's Res. Hosp., Memphis, TN

Abstract: Schizophrenia is a multifaceted neurodevelopmental disorder characterized by notoriously intractable positive, cognitive, and negative symptom categories. The severity of the negative symptom amotivation is negatively correlated with social and occupational functioning and is not currently met with effective treatment options. Advances in treatment are hampered by a lack of a mechanistic understanding of the neural circuits underlying this symptom. Despite being implicated in motivational states, the dorsal striatum is largely overlooked when studying

motivated behavior. Further, disruptions in thalamic nuclei are involved in all facets of schizophrenia symptomology, but the role of the thalamus in amotivation remains unknown. Here, we use a mouse model of 22q11 deletion syndrome (22q11DS), a genetic disorder in which 30% of individuals develop symptoms that are indistinguishable from idiopathic schizophrenia, to test the role of the thalamostriatal pathway in motivated behavior. Using the progressive ratio behavior task and *ex vivo* whole-cell patch-clamp electrophysiology and optogenetics, we show that these mice display amotivation and exhibit weakened excitatory inputs to the dorsomedial striatum (DMS) from the parafascicular nucleus of the thalamus (Pf). Using chemogenetics in wildtype mice, we show that mimicking Pf-DMS weakening *in vivo* recapitulates the amotivation phenotype, causally implicating this circuit in amotivational states. Striatal cholinergic interneurons (CHIs) modulate Pf signaling onto the principal medium spiny neurons of the DMS and provide a basal cholinergic tone in the DMS by firing tonically. We show that, compared to wildtype mice, there are substantially more spontaneously active CHIs in 22q11DS mice. Moreover, blocking signaling through acetylcholine receptors rescues the weakened Pf-DMS drive. These findings are the first to implicate the Pf-DMS pathway in amotivation symptoms arising in 22q11DS and point to intra-striatal acetylcholine signaling as a possible mechanistic linchpin in the etiology of amotivation.

Disclosures: **M.H. Patton:** None. **B.J.W. Teubner:** None. **S.S. Zakharenko:** None.

Poster

PSTR177. Neuropathology, Genetics, Genomics, and Molecular Mechanisms: Schizophrenia

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR177.23/UU28

Topic: G.08. Other Psychiatric Disorders

Title: Non-clinical Profile of a Novel Muscarinic M4 Receptor Positive Allosteric Modulator in Animal Models of Schizophrenia

Authors: A. SHINDE, A. MOHAMMED, R. SUBRAMANIAN, *J. THENTU, V. BENADE, V. PALACHARLA, R. MEDAPATI, R. KALLEPALLI, R. ABRAHAM, K. BOJJA, A. SHAIKH, R. NIROGI;
Suven Life Sci. Ltd., Hyderabad, India

Abstract: Centrally acting muscarinic acetylcholine receptor antagonists like atropine and scopolamine can induce psychosis like symptoms. Xanomeline, a muscarinic M1/M4 preferring agonist attenuated the effects of amphetamine (animal model for schizophrenia) in the wild type mice, however such effects were absent in muscarinic M4 knockout mice. In addition, xanomeline was also found to be effective in attenuating disease symptoms in schizophrenic patients. Thus, muscarinic M4 agents can modulate brain circuitry that is dysregulated in schizophrenia. SUVN-L8203032 is a novel, potent and selective positive allosteric modulator of muscarinic M4 receptors. SUVN- L8203032 showed good oral bioavailability and brain

penetration at the dose of 3 mg/kg. SUVN-L8203032 was assessed for the receptor occupancy of the allosteric site of muscarinic M4 receptors in the brain. SUVN-L8203032 showed dose dependent receptor occupancy at doses of 3, 10 & 20 mg/kg. SUVN-L8203032 was assessed for its effects on amphetamine induced hyperlocomotion in rats at doses of 3, 10 & 20 mg/kg. At the tested doses, SUVN-L8203032 dose dependently attenuated amphetamine-induced hyperlocomotion. The observation from the amphetamine-induced hyperlocomotion assay correlated well with the occupancy at the allosteric site of muscarinic M4 receptors. SUVN-L8203032 is being further characterized in animal models for positive and cognitive symptoms of schizophrenia. SUVN-L8203032 could be a promising agent for targeting positive and cognitive symptoms of schizophrenia.

Disclosures: **A. Shinde:** A. Employment/Salary (full or part-time); Suven Life Sciences Ltd. **A. Mohammed:** A. Employment/Salary (full or part-time); Suven Life Sciences Ltd. **R. Subramanian:** A. Employment/Salary (full or part-time); Suven Life Sciences Ltd. **J. Thentu:** A. Employment/Salary (full or part-time); Suven Life Sciences Ltd. **V. Benade:** A. Employment/Salary (full or part-time); Suven Life Sciences Ltd. **V. Palacharla:** A. Employment/Salary (full or part-time); Suven Life Sciences Ltd. **R. Medapati:** A. Employment/Salary (full or part-time); Suven Life Sciences Ltd. **R. kallepalli:** A. Employment/Salary (full or part-time); Suven Life Sciences Ltd. **R. Abraham:** A. Employment/Salary (full or part-time); Suven Life Sciences Ltd. **K. Bojja:** A. Employment/Salary (full or part-time); Suven Life Sciences Ltd. **A. Shaikh:** A. Employment/Salary (full or part-time); Suven Life Sciences Ltd. **R. Nirogi:** A. Employment/Salary (full or part-time); Suven Life Sciences Ltd.

Poster

PSTR177. Neuropathology, Genetics, Genomics, and Molecular Mechanisms: Schizophrenia

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR177.24/VV1

Topic: G.08. Other Psychiatric Disorders

Support: National Research Foundation of Korea Grant NRF-2021R1C1C1012901

Title: Identification of schizophrenia using an interpretable radiomics model with structural magnetic resonance imaging of the cerebellum

Authors: *M. BANG[1], Y. PARK[2], S.-H. LEE[1];
[1]CHA Bundang Med. Ctr., Seongnam, Korea, Republic of; [2]Radiology, Yonsei Univ. Col. of Med., Seoul, Korea, Republic of

Abstract: Background: The cerebellum is involved in higher-order cognitive and affective processing as well as sensorimotor functions. Although structural abnormalities in the cerebellum have been demonstrated in patients with schizophrenia, neuroimaging techniques are

not yet applicable to identify schizophrenia due to the subtle nature of brain pathogenesis. Radiomics quantifies meaningful “hidden” information within medical images by computing high-dimensional features related to the shape and texture information. We aimed to develop a robust diagnostic model for schizophrenia using radiomics features extracted from T1-weighted (T1) magnetic resonance images of the cerebellum.
Methods: A total of 347 participants (180 with schizophrenia and 167 healthy controls [HCs]) were allocated to training (126 with schizophrenia and 120 HCs) and test (54 with schizophrenia and 47 HCs) sets. We obtained 2,782 radiomic features of the 13 cerebellar subregions from T1 images. Following feature selection with mutual information, a histogram-based gradient boosting classifier was trained. Discrimination and calibration of the model were evaluated to assess model performance. The model was validated on the test set. SHapley Additive exPlanations (SHAP) was applied to explore the interpretability of the model.
Results: We identified 20 first- and second-order radiomic features to differentiate participants with schizophrenia from HCs. In the test set, the radiomics model had an area under the curve, accuracy, sensitivity, and specificity of 0.82 (95% confidence interval: 0.74-0.90), 76.2%, 75.9%, and 78.8%, respectively. The calibration plot showed a good agreement between the predicted and actual rates of schizophrenia (Brier score = 0.22). The model explanation by SHAP suggested that second-order features from the left lobule VIIIa, right lobule IX, and right lobule VIIa were highly associated with the risk of schizophrenia.
Discussion: The cerebellum-focused radiomics model shows its robustness for the diagnosis of schizophrenia. Our results suggest that microstructural alterations in the posterior cerebellum are disease-defining features of schizophrenia. Radiomic features may be a potential source of biomarkers to support the objective diagnosis and discover novel treatment targets for schizophrenia.

Disclosures: **M. Bang:** None. **Y. Park:** None. **S. Lee:** None.

Poster

PSTR177. Neuropathology, Genetics, Genomics, and Molecular Mechanisms: Schizophrenia

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR177.25/VV2

Topic: G.08. Other Psychiatric Disorders

Support: KHIDI Grant HI17C0870
NRF Grant 2019M3C7A1030625

Title: Identifying clinical and proteomic markers for early diagnosis and prognosis prediction of major psychiatric disorders- a 48 month follow-up study

Authors: ***H. LEE**[1], D. HAN[1], S. RHEE[1], J. LEE[2], J. KIM[1], Y. LEE[3], E. KIM[4], D. PARK[5], S. ROH[6], M. BAIK[7], H. JUNG[8], T. LEE[9], M. KIM[1], H. KIM[1], S. KIM[1], J. KWON[1], Y. AHN[1], K. HA[10];

[1]Seoul Natl. Univ. Hosp., Seoul, Korea, Republic of; [2]Uijeongbu Eulji Med. Ctr., Uijeongbu, Korea, Republic of; [3]Kosin Univ. Gospel Hosp., Busan, Korea, Republic of; [4]Seoul Natl. Univ. Col. of Med., Seoul, Korea, Republic of; [5]Natl. Ctr. for Mental Hlth., Seoul, Korea, Republic of; [6]Hanyang Univ. Hosp., Seoul, Korea, Republic of; [7]Kyung Hee Univ. Med. Ctr., Seoul, Korea, Republic of; [8]SMG-SNU Boramae Med. Ctr., Seoul, Korea, Republic of; [9]Pusan Natl. Univ. Yangsan Hosp., Busan, Korea, Republic of; [10]Univ. of British Columbia, Vancouver, BC, Canada

Abstract: Background: Biological markers, particularly levels of blood proteins, could serve as crucial tools for predicting disease progression in individuals with a high risk of developing psychiatric disorders. This study aimed to identify clinical and blood protein markers that have disease prediction and progression monitoring potential. **Methods:** We recruited 90 individuals at clinical high risk (CHR) of major psychiatric disorders, including psychosis, bipolar, and depression, and tracked their progression for up to 4 years. Three predictive models were used—Model 1 (100 clinical variables), Model 2 (158 peptides), and Model 3 (100 clinical variables + 158 peptides). The models were evaluated using the area under the receiver operating characteristic (AUROC) values. Linear mixed-effect analysis was used to observe distinct patterns of the selected clinical and protein features for 12 and 24 months among the patients who did (CHR-T) and did not transition (CHR-NT) to the disease state and those in the extended risk group (patient group). **Result:** Eighteen individuals at CHR of major psychiatric disorders developed the disorders over an average of 17.7 months. The combined model showed the highest discriminatory performance (AUROC = 0.73). Among the protein markers, MDHC and TAGL2 levels were lesser in the CHR-T compared to that in the CHR-NT group. Levels of four proteins (CO9, ITIH4, VWF, and CRP) were decreased in the patient compared to that in the CHR-NT group. **Conclusion:** We identified the baseline clinical and protein markers that could aid in prediction of individuals at a high risk of developing psychiatric disorders.

Disclosures: H. Lee: None. **D. Han:** None. **S. Rhee:** None. **J. Lee:** None. **J. Kim:** None. **Y. Lee:** None. **E. Kim:** None. **D. Park:** None. **S. Roh:** None. **M. Baik:** None. **H. Jung:** None. **T. Lee:** None. **M. Kim:** None. **H. Kim:** None. **S. Kim:** None. **J. Kwon:** None. **Y. Ahn:** None. **K. Ha:** None.

Poster

PSTR177. Neuropathology, Genetics, Genomics, and Molecular Mechanisms: Schizophrenia

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR177.26/VV3

Topic: G.08. Other Psychiatric Disorders

Title: Whole-brain structural and functional neuroimaging correlates of set-shifting across different mental disorders: a coordinate-based meta-analysis

Authors: *N. MEDA, R. CAZZARO, M. ROMANELLI, F. SAMBATARO;
Univ. di Padova, Padova, Italy

Abstract: Set-shifting skills allow individuals to switch flexibly from a behavior to a more adaptive one in the face of negative feedback from the environment. This complex ability, which is part of cognitive control functions, is impaired in several mental and neurodevelopmental disorders, such as schizophrenia (SCZ), obsessive-compulsive disorder (OCD) and anorexia nervosa (AN). Although neuroimaging studies on healthy adults have highlighted the activity of frontal and parietal cortices as the neural bases of stop-and-switch behavior, it is unclear whether the neural correlates of set-shift impairment are shared or not between different mental disorders.Therefore, we systematically screened the neuroimaging literature and conducted a coordinate-based meta-analysis (CBMA) to determine whether the neuroimaging features of set-shifting impairment are common to different mental disorders.Out of 1932 publications screened, we meta-analysed 22 functional neuroimaging studies that investigated the neural activity differences between healthy subjects (n=447; Mean female percentage = 42) and people with a mental disorder (n=467; mF% = 42; SCZ, major depressive disorder, autism, generalized anxiety disorder, ADHD, AN, OCD) during a set-shifting paradigm on the scan.We found that three brain clusters were more active in patients (with schizophrenia, autism spectrum disorders or generalized anxiety disorder) than healthy subjects during set-shifting with respect to baseline. The clusters were localized in the right medial frontal/anterior cingulate gyrus, the right superior parietal lobule and the left superior temporal gyrus. No brain clusters of hypoactivation were found.The meta-analytical evidence herein identifies brain hubs belonging to the frontoparietal network (FPN) as putative regions of altered activity in some mental disorders during set-shifting. Given that the lateral regions of the FPN are recruited in healthy subjects when cognitive flexibility is required, the findings of this meta-analysis support the view of altered activity of normally recruited regions in patients applying their set-shifting abilities. Taking into account the evidence that FPN subregions act as neural substrates of an error detection network, these findings suggest that patients need to hyperactivate these brain regions to update their information in the face of negative feedback from the environment, and failure to do so would lead to inflexible behavior.

Disclosures: N. Meda: None. **R. Cazzaro:** None. **M. Romanelli:** None. **F. Sambataro:** None.

Poster

PSTR178. Genetic Techniques to Target and/or Manipulate Cells

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR178.01/VV4

Topic: I.01. Molecular, Biochemical, and Genetic Techniques

Support: R01-NS109916

Title: Transcriptional Regulation of Postnatal Neuronal Maturation by the KLF family of Transcription Factors

Authors: *R. KIRK[1], L. SUN[1], E. XIAO[1], S. B. NELSON[2];
[2]Dept Biol MS#008, [1]Brandeis Univ., Waltham, MA

Abstract: During the first month of postnatal development, neurons in the mouse cortex undergo dramatic changes in their gene expression, morphology, and physiology that are crucial for establishing balanced synaptic transmission in the mature brain. As they mature, cortical neurons globally rebalance the repertoire of ion channels and neurotransmitter receptors they express, yet the transcriptional regulatory mechanisms underlying this postnatal transition remain poorly understood. We sought to identify transcription factors that mediate the rebalancing of ion channel subunits by analyzing RNA-seq and ATAC-seq data obtained at the beginning (P2) and end (P30) of the first month of postnatal development. This identified the Kruppel-like Factor (KLF) family of transcription factors as putative regulators of the postnatal change in ion channel gene expression. Specifically, we observed a decline in the expression of the transcriptional activators *Klf6* and *Klf7* concomitant with rising expression of the transcriptional repressors *Klf9* and *Klf13* during the first 4 postnatal weeks. We hypothesized that this shift from activating to repressive KLF's mediates the repression of the immature gene expression program in mature neurons. To test our predictions, we developed an in vivo CRISPR-interference (CRISPRi)-based knockdown strategy to knock down members of either transcription factor family individually or in combination with the family member of shared regulatory valence (Klf6/Klf7 or Klf9/Klf13). By injecting neonatal Emx1-Cre;LSL-dCas9-KRAB mice with AAV9 containing guide RNA's (gRNA) targeting our genes of interest, we are able to reliably knock down multiple genes by >95% using a single virus. Upon sequencing the RNA of infected cells isolated by FACS at P18-P20, we identified the cell-autonomous effects of individual or combinatorial transcription factor knockdown(s) on the neuronal transcriptome. These experiments demonstrated that Klf9 and Klf13 are partially compensatory transcription factors that cooperatively repress a set of genes associated with the cytoskeleton and some ion channel genes. However, we found no effect on synaptic or intrinsic electrophysiological properties of neurons lacking Klf9/13 at P18. Further experiments will examine how the loss of Klf9/13 impacts maturation of cortical neurons across early postnatal development and investigate whether Klf6 & Klf7 act on the same targets as the repressive KLF's. Together, our results offer novel examples of redundancy and synergy in transcription factor function while demonstrating the utility of CRISPRi for dissecting transcriptional regulatory networks in vivo.

Disclosures: R. Kirk: None. **L. Sun:** None. **E. Xiao:** None. **S.B. Nelson:** None.

Poster

PSTR178. Genetic Techniques to Target and/or Manipulate Cells

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR178.02/VV5

Topic: I.01. Molecular, Biochemical, and Genetic Techniques

Support: NIH Grant R03AG063264
NIH Grant R21NS109918

NIH Grant R01DA048815
NIH Grant R01DA048815-03S1

Title: A novel FlpO recombinase mouse line to target a subset of reactive astrocytes

Authors: *T. UBINA[1], S. SRIRAM[5], E. CONTRERAS[5], W. AGNEW-SVOBODA[5], E. WILSON[6], V. SANTHAKUMAR[2], T. FIACCO[3], M. RICCOMAGNO[4];
[2]Mol. Cell and Systems Biol., [3]Cell Biol. & Neurosci., [4]Molecular, Cell and Developmental Biol., [1]Univ. of California, Riverside, Riverside, CA; [5]Univ. of California Riverside, Riverside, CA; [6]Univ. of California, Riverside Biomed. Sci. Grad. Program, Riverside, CA

Abstract: Astrocytes ordinarily execute vital processes for proper brain function, but reprogram into "reactive" astrocytes (RAs) following injury. Astrocytes become reactive to varying degrees in virtually all neurodegenerative diseases and brain disorders. When astrocytes become reactive, they undergo changes in gene expression, morphology and function. This response varies based on the type of insult and brain region that is affected. With the aim of targeting, labeling and manipulating RAs, we have begun to design RA-specific recombinase lines. Our lab has previously generated an *Lcn2-CreER^{T2}* line that is capable of permanently labeling a subset of RAs and other reactive cells. Here, we describe the generation of a different recombinase line driven by the endogenous Complement 3 (C3) promoter. C3 is involved in the complement cascade and has been shown to be significantly upregulated in a subset of RAs. We used this knowledge to develop an inducible FlpO knock-in mouse line (*C3-FlpOER^{T2}*) that targets a subset of RAs. We then characterized the *C3-FlpOER^{T2}* line in several models of neuroinflammation. The generation of a FlpO line opens up the possibility of combinatorial approaches using well validated astrocytic Cre lines to study RA function and heterogeneity.

Disclosures: T. Ubina: None. **S. Sriram:** None. **E. Contreras:** None. **W. Agnew-Svoboda:** None. **E. Wilson:** None. **V. Santhakumar:** None. **T. Fiacco:** None. **M. Riccomagno:** None.

Poster

PSTR178. Genetic Techniques to Target and/or Manipulate Cells

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR178.03/VV6

Topic: I.01. Molecular, Biochemical, and Genetic Techniques

Support: NIH Grant U19MH114830

Title: Transgenic tools for Cell Type specific Intersectional Targeting of Genetically Encoded Sensors and Effectors.

Authors: *S. NARAYAN[1], T. L. DAIGLE[1], J. BENDRICK[1], D. BIRMAN[2], Z. YE[2], P. A. GROBLEWSKI[1], P. BALARAM[1], S. WAY[1], K. RONELLENFITCH[1], N. A. STEINMETZ[2], H. ZENG[1], B. TASIC[1];
[1]Allen Inst., Seattle, WA; [2]Univ. of Washington, Seattle, WA

Abstract: The advent of single cell genomics has given us the ability to delineate cell types based on gene expression patterns and thus enabled the creation of driver lines for intersectional targeting of specific cell populations. When coupled with suitable reporters, these driver lines give us unprecedented access to specific cell populations involved in complex behaviors. Cell types are rarely defined by unique marker genes. Therefore, tools based on intersectional expression of multiple genes are critical for cell-type specific access. We developed a transgenic platform using a new landing pad mouse ES cell line for Bxb1-integrase-mediated cassette exchange at the TIGRE locus. We called this platform TIGRE 3.0. The overall stability and tight regulation of reporter genes at this locus allows for the insertion of complex reporter constructs that can integrate input from multiple recombinases thus allowing for a finer control of cell type access. Here we describe three dual-recombinase responsive (Cre/FLP-dependent), reporter lines for intersectional targeting of cells in the mouse brain. These lines, Ai195 and Ai210 both express the Calcium indicator jGCaMP7f and jGCaMP7s respectively whereas Ai211 expresses the red shifted opsin, Chrimson, under Cre and Flp control. These reporters, when used with existing viral or transgenic targeting strategies, enable cell type specific intersectional targeting experiments at an unprecedented level of precision.

Disclosures: S. Narayan: None. **T.L. Daigle:** None. **J. Bendrick:** None. **D. Birman:** None. **Z. Ye:** None. **P.A. Groblewski:** None. **P. Balaram:** None. **S. Way:** None. **K. Ronellenfitch:** None. **N.A. Steinmetz:** None. **H. Zeng:** None. **B. Tasic:** None.

Poster

PSTR178. Genetic Techniques to Target and/or Manipulate Cells

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR178.04/VV7

Topic: I.01. Molecular, Biochemical, and Genetic Techniques

Title: Polymer-based films facilitate single-step targeted expression of genetically-encoded activity sensors for in vivo microendoscopic calcium imaging

Authors: D. CHENG[1], C. JONS[2], E. APPEL[2], ***J. NASSI**[1];
[1]Inscopix, Inc., Mountain View, CA; [2]Materials Sci. and Engin., Stanford Univ., Stanford, CA

Abstract: Microendoscopic calcium imaging with miniature microscopes is a powerful approach for studying the relationship between neural activity and behavior in freely behaving animals. The technique relies on the use of genetically-encoded calcium indicators, most commonly GCaMP, to monitor neural activity in specific cell populations. Adeno-associated virus (AAV) injections are a widely utilized method of expressing GCaMP in target brain regions. However, following an AAV injection, a second surgical step is often needed for a gradient refractive index (GRIN) lens to be implanted to enable optical access and monitor fluorescence deep in the brain. The two-step procedure can reduce experimental success rates due to difficulties in targeting both the virus and the lens to the same site. This, in turn, limits throughput and scale for translational and preclinical studies. Here, we engineered a novel polymer coating that can be

applied to the surface of a GRIN lens to deliver AAVs to the brain. This reduces the number of required surgeries to one and guarantees alignment between GCaMP expression and lens in the brain. To do so, we screened a library of polysaccharides of varied molecular weights to identify polymers that provide diffusion kinetics appropriate to slow the release of virus. Bench assays were developed and implemented to further screen the most promising polymer candidates based on their release kinetics, swelling behavior, biocompatibility and ability to stabilize virus during storage. Lead candidates were then further screened in-vivo, by implanting GRIN lenses coated with AAV-GCaMP polymer films into the dorsal medial striatum and prefrontal cortex of mice and rats. These in-vivo experiments demonstrated that the polymer coated lenses consistently provide high cell count fields of view with calcium transient kinetics and signal-to-noise ratio supporting high quality in vivo calcium imaging. Critically, by engineering a polymer coating that slows viral diffusional release, we increased lens implantation working time and expanded capabilities to image deeper brain regions in additional species. These capabilities represent an important technical advance that promises to accelerate breakthrough discoveries and enable higher throughput and scale for translational and preclinical neuroscience research.

Disclosures: **D. Cheng:** A. Employment/Salary (full or part-time); Inscopix, Inc. **C. Jons:** B. Contracted Research/Research Grant (principal investigator for a drug study, collaborator or consultant and pending and current grants). If you are a PI for a drug study, report that research relationship even if those funds come to an institution.; Inscopix, Inc. **E. Appel:** B. Contracted Research/Research Grant (principal investigator for a drug study, collaborator or consultant and pending and current grants). If you are a PI for a drug study, report that research relationship even if those funds come to an institution.; Inscopix, Inc. **J. Nassi:** A. Employment/Salary (full or part-time); Inscopix, Inc.

Poster

PSTR178. Genetic Techniques to Target and/or Manipulate Cells

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR178.05/VV8

Topic: I.01. Molecular, Biochemical, and Genetic Techniques

Support: UVA Brain Institute

Title: FLEX2: novel Cre-dependent AAVs with reduced off-target expression

Authors: ***E. PEREZ-REYES**[1], M. J. FAILOR[2], J. M. SAN PIETRO[2], L. ERGUN[2], K. SINGH[2], R. P. GAYKEMA[3];
[1]Pharmacol., Univ. of Virgina, Charlottesvle, VA; [2]Univ. of Virginia, Charlottesville, VA; [3]Pharmacol., Sch. of Med., Charlottesville, VA

Abstract: Key tools for studying neural circuits combine Cre recombinase dependent expression of either markers (GFP), optogenetic and chemogenetic actuators (e.g. channelrhodopsin and DREADDs) or biosensors (GCaMPs). A common design is to use AAVs to deliver the gene-of-

interest that is made Cre dependent by flanking its inverted open-reading frame with specific Cre recognition sites (loxP/lox2272; DIO or FLEX design). *Recent studies have sounded the alarm on leaky, non-Cre dependent expression.* One approach is to inject these AAVs into wild-type C57BL/6 mice. Using this approach we find *6% off-target expression* of FLEX-tdTomato (Addgene 28306). This level of off-target expression of FLEX/DIO AAVs leads to confounds in the interpretation of results, and coupled with their wide-spread use (>2,000 papers), highlights the need for improved Cre-dependent designs. Fixing the off-target leak begins with understanding two of the main sources of the problem. *Problem #1* is that current FLEX sequences recombine during cloning, plasmid amplification, and AAV production (Fischer et al., 2019). To reduce recombination we replaced the lox sites with mutant sites that form shorter hairpins (lox71/66). In vitro testing of this FLEX2 design indicates spontaneous recombination is reduced 4-fold and expression after Cre recombination is improved 8-fold. *Problem #2* is that the AAV ITR sequences have promotor activity (Haberman et al., 2000), which leads to the expression of the transgene even though it is oriented 3'-5' relative to the neuronal promoter. Our solution was to split both the open-reading frame of the transgene and a synthetic intron into two fragments, and then invert one of them. The split intron serves to remove residual lox sites from the final coding sequence. Preliminary data shows the this approach rescues the ability to make AAV encoding a toxic protein, diphtheria toxin A chain. In vivo testing of our FLEX2 split-GFP in C57BL/6J mice showed no detectable GFP fluorescence. However, a low level of GFP could be detected after antibody amplification. *When compared to a current FLEX-GFP design, FLEX split-GFP demonstrated a 15-fold reduction in off-target expression.*

Disclosures: E. Perez-Reyes: None. **M.J. Failor:** None. **J.M. San Pietro:** None. **L. Ergun:** None. **K. Singh:** None. **R.P. Gaykema:** None.

Poster

PSTR178. Genetic Techniques to Target and/or Manipulate Cells

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR178.06/VV9

Topic: I.01. Molecular, Biochemical, and Genetic Techniques

Support: NIH Grant R15HL165397
NIH Grant P20GM103408
NIH Grant P20GM109095

Title: Vascular compartment labeling by intravenous rAAV9 results in cell lineage tracing to diverse parenchymal cells in adult mice

Authors: *B. MORRISON, **A. PUGEL**, A. SOTO-AVELLANEDA, J. HOLMES, A. SCHOENFELD, S. ALSAIFI, G. KOIRALA, A. OXFORD;
Dept. of Biol. Sci., Boise State Univ., Boise, ID

Abstract: A growing number of studies have reported the transduction of a myriad of parenchymal cell types following an intravenous recombinant adeno-associated virus (rAAV) injection. This finding has significant implications for potential routes of minimally invasive human gene therapy. However, to date, an empirically backed rationale for any mechanism underlying this extravascular transgene expression is lacking. What's more, the literature is fragmented, with studies focusing on a single cell type or tissue that is often within the context of a disease, thereby confounding a broader interpretation of these results. We sought to build upon our previous study, which focused on the notion of endothelial cell contributions via transdifferentiation or extracellular vesicles to transgene expression by parenchymal cells. This previous work employed tamoxifen-induced cell lineage tracing in adult transgenic mice using the vascular endothelial cadherin (VEcad) promoter to drive reporter expression. Since VEcad has been shown to be expressed by very rare non-endothelial cell types, we initiated cell lineage tracing in this study specifically within the vascular compartment by intravenous delivery of rAAV serotype 9 (rAAV9), a variant shown to readily transduce endothelial cells. We also utilized multiple time points that matched our previous study for direct comparison in addition to examining 25 tissues and cell types in a context that did not involve pathogenic induction so that broad interpretations might be more readily possible. We found numerous cell types traced from intravenous rAAV9, including hippocampal neurons and Purkinje cells, all of which have been previously reported. However, importantly, we also observed that five cell types (including skeletal myocytes, pancreatic beta cells, pancreatic acinar cells, duodenal epithelial cells and ileal epithelial cells) traced from both intravenous rAAV9 and the VEcad promoter from our previous study. Thus, these five cell types warrant further investigation as potential downstream targets of endothelial cell transdifferentiation or extracellular vesicles *in vivo*.

Disclosures: B. Morrison: None. **A. Pugel:** None. **A. Soto-Avellaneda:** None. **J. Holmes:** None. **A. Schoenfeld:** None. **S. Alsaifi:** None. **G. Koirala:** None. **A. Oxford:** None.

Poster

PSTR178. Genetic Techniques to Target and/or Manipulate Cells

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR178.07/VV10

Topic: I.01. Molecular, Biochemical, and Genetic Techniques

Support: Aligning Science Across Parkinson's (ASAP-020495) through the Michael J. Fox Foundation for Parkinson's Research (MJFF)

Title: Development of Systemic rAAV Tools for Regenerative Spiny Mice

Authors: *A. CHUNG[1], R. R. DONAHUE[2], C. LIN[1], X. CHEN[1], M. ZHANG[1], A. SEIFERT[2], V. GRADINARU[1];
[1]Biol. and Biol. Engin., Caltech, Pasadena, CA; [2]Biol., Univ. of Kentucky, Lexington, KY

Abstract: Spiny mice (genus *Acomys*) demonstrate a remarkable capability to repair injuries to the central nervous system (CNS) and peripheral organs, and understanding this process could inform future therapeutic approaches for neurodegenerative disorders such as Parkinson's disease. Work to date in spiny mice has utilized direct injection of recombinant adeno-associated viruses (rAAVs) into the CNS for neural circuit tracing and gene editing in the brain. However, to develop neural disease models and broaden our understanding of central and peripheral nerve regeneration in the spiny mouse, we need non-invasive systemic gene delivery vectors similar to those we have developed for *Mus musculus*. Here, we characterized the transduction profiles of engineered rAAVs in the CNS and peripheral nervous system (PNS) of spiny mice following systemic administration. We administered a pool of AAV9 variants (AAV9, PHP.eB, CAP-B10, CAP-B22, CAP-Mac, X1.1, MaCPNS1, and MaCPNS2), each carrying a unique barcoded cargo, to spiny mice via the intraperitoneal route. Next-generation sequencing (NGS) of the barcode sequences revealed that MaCPNS1 was highly enriched in the CNS, small intestine, and large intestine. We further characterized MaCPNS1 by using it to deliver an eGFP cargo driven by the ubiquitous CAG promoter via the retro-orbital sinus. We found that MaCPNS1 transduced neurons and astrocytes in the brain of the spiny mouse. Further, we found robust transduction of the PNS in the dorsal root ganglia, neurons in the small and large intestines, and nerve fibers in the heart and ear. Together, our results demonstrate that MaCPNS1 can serve as a strong basis for an rAAV toolkit for accessing the nervous system of spiny mice and delivering functional cargo, enabling the development of models of neurodegenerative diseases in the spiny mice, and investigation of the role of the nervous system in regeneration.

Disclosures: **A. Chung:** None. **R.R. Donahue:** None. **C. Lin:** None. **X. Chen:** None. **M. Zhang:** None. **A. Seifert:** None. **V. Gradinaru:** None.

Poster

PSTR178. Genetic Techniques to Target and/or Manipulate Cells

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR178.08/VV11

Topic: I.01. Molecular, Biochemical, and Genetic Techniques

Support: NIH Grant DC016905
Hearing Health Foundation
National Ataxia Foundation

Title: Comparative analysis of transduction efficiency, viral spread, tropism, and axonal transport of six adeno-associated viral vector serotypes in mouse inferior colliculus and cerebellum

Authors: I. F. WITTEVEEN, ***T. S. BALMER**;
Sch. of Life Sci., Arizona State Univ., Tempe, AZ

Abstract: Adeno-associated virus (AAV) vectors have emerged as a widely utilized gene delivery tool in neuroscience research and are becoming more common in clinical settings. Naturally occurring and engineered serotypes have varying abilities to infect the brain and express transgenes. AAV transduction efficiency in specific neuron types is unpredictable, so an empirical approach is often required to determine the best serotype for each population of cells. Previous studies have investigated variations in transduction efficacy and tropism of different AAV serotypes in some brain regions including the striatum, cerebral cortex, and hippocampus, and have been a valuable contribution to research in these areas. AAV serotype transduction and tropism in the inferior colliculus and cerebellum of the mouse brain have not been examined. Validating AAVs for use in these brain regions could accelerate the study of auditory and motor circuits and their associated disorders. Here we report a comprehensive comparison of the AAV serotypes 1, 2, 5, 8, 9, and the directed evolution derived AAVrg, in the inferior colliculus and cerebellum. The AAVs were identical apart from their different serotypes, each having a synapsin promotor and expressing GFP (AAV-hSyn-GFP). Identical titers and volumes were injected sterotaxically into the inferior colliculus and cerebellum of male and female C57BL6 mice. After 2 weeks, the brains were sectioned and imaged. Transduction efficacy and viral spread was evaluated through fluorescence analysis, and tropism was assessed by analyzing cell morphology and axonal projections to known targets. Retrograde labeling was also characterized. In both the cerebellum and inferior colliculus, AAV1 expressed GFP in a larger volume than all other serotypes, indicating higher viral spread. AAV1 also produced significantly brighter labeling than all other serotypes in the cerebellum and significantly brighter labeling than AAV2, AAV5, and AAVrg in the inferior colliculus, indicating superior transgene expression. These results will help researchers choose the most appropriate AAV serotype to express genes in these brain regions.

Disclosures: I.F. Witteveen: None. **T.S. Balmer:** None.

Poster

PSTR178. Genetic Techniques to Target and/or Manipulate Cells

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR178.09/VV12

Topic: I.01. Molecular, Biochemical, and Genetic Techniques

Support: IR-FG24932

Title: A recently developed "microglia-targeting" AAV capsid enables specific genetic access to hippocampal and neocortical excitatory neurons

Authors: *W. CAO, K. REPMAN, X. XU;
Anat. and Neurobio., Univ. of California, Irvine, Irvine, CA

Abstract: Genetically modified adeno-associated viruses (AAV) using enhancer elements and capsid variants have been used for cell-type specific targeting in the central nervous system. It

has been recently reported that adeno-associated virus (AAV) capsid variants (AAV-MG1.1 and AAV-MG1.2) produced by directed evolution capsid engineering (Lin et al., 2022), can mediate efficient in vitro and in vivo microglial transduction, capable of delivering various genetic payloads into microglia with high efficiency. In this study we report that AAV-MG1.2 actually enables specific genetic access to hippocampal and neocortical excitatory neurons in vivo, but does not infect non-neuronal cells including microglia in vivo. We packaged CAG-EGFP and CAG-tdTomato into the AAV capsid MG1.2, respectively, and stereotaxically injected the virus into hippocampal CA1, subiculum and visual cortical regions of adult wild type mice. Through quantification of CaMKIIa+ and GABA- immunostaining, we identified almost 100% of EGFP- or tdTomato-expressing cells to be excitatory cells in hippocampal CA1 / subiculum, and identified 90 % of EGFP- or tdTomato-expressing cells to be excitatory cells in visual cortex. Thus the MG1.2 capsid primarily labeled excitatory principal neurons in hippocampal CA1/subiculum and visual cortex. In addition, we found that the MG1.2 capsid specifically labeled the deep layer of the CA1 pyramidal layer in a titer-dependent manner. Lower virus titers ($5.15x10^{12}$ GC/ml compared to $5.15x10^{13}$ GC/ml for MG1.2-CAG-eGFP) resulted in more precise labeling specifically within the deep layer. This specificity for the deep layer was more pronounced in the ventral CA1. Given the cell type heterogeneity among CA1 pyramidal cells exists along the superficial-deep axis, AAV-MG1.2 can be used to genetically target the deep sublayer of CA1 stratum pyramidale for structural and functional analysis. Taken together our new discovery regarding the AAV capsid MG1.2 expands our genetic toolset to target overall and specific excitatory cell types in the brain.

Disclosures: **W. Cao:** None. **K. Repman:** None. **X. Xu:** None.

Poster

PSTR178. Genetic Techniques to Target and/or Manipulate Cells

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR178.10/VV13

Topic: I.01. Molecular, Biochemical, and Genetic Techniques

Support: NIH Grant R56 AG078565
NIH Grant R01 NS089586
Shriners Hospitals for Children 87530-PHI-22

Title: Cell-specific AAV-mediated transgene expression in oligodendrocytes

Authors: *N. ZUBIN, J. COAKLEY, T. CAMPION, S. KANG;
Dept. of Neural Sci., Temple Univ., Philadelphia, PA

Abstract: Recombinant adeno-associated viral vectors (AAVs) are a powerful tool for gene delivery to various cells *in vitro* and *in vivo.* It is widely used in neuroscience research to interrogate cellular and neurocircuit functions and map brain connectivity. Despite their successful use for a wide range of neurons and astrocytes, other non-neuronal cells are known to

be refractory to or have poor target cell specificity with the available AAV serotypes. Oligodendrocytes are the myelin-forming glia in the central nervous system (CNS), crucial for rapid axonal conduction and metabolic support to axons. Their adaptative generation according to neuronal activity, maintenance, dysfunctions, and regeneration are critical factors for adult CNS function. In recent years, oligodendrocyte-targeted AAV transduction has been improved either by adopting a short (~2 kb) gene promoter of a myelin protein (e.g., MBP or MAG) or by developing a new serotype with oligodendrocyte-preferred tropism. However, even in those settings, significant off-target transgene expression in transduced cells may confound study results concerning mechanisms of intercellular interactions among oligodendrocytes and neurons or other glia. To develop a better AAV system with improved oligodendrocyte specificity, we employed the Cre-loxP-dependent flip-excision (FLEx) switch in combination with Olig001, a capsid with oligodendrocyte-preferred tropism. We assessed nuclear fluorescent reporter protein expression after AAV-mediated transduction of the brain, confirming that AAV-delivered transgenes are expressed under the control of the CAG promoter only in Cre-active oligodendrocytes. In addition, we characterized distinct advantages of 3 different Cre lines for the transduction efficiency or cell specificity in AAV-mediated transgene expression. This approach was further applied to express diphtheria toxin a-chain (DTa) in the corpus callosum or hippocampus, serving as a means for inducing focal demyelination in gray and white matter brain areas. Our efforts established an advanced AAV-mediated transgene delivery method targeting oligodendrocytes and demonstrated its usefulness for future preclinical studies that model oligodendrocyte-related disease states and seek to identify underlying mechanisms.

Disclosures: **N. Zubin:** None. **J. Coakley:** None. **T. Campion:** None. **S. Kang:** None.

Poster

PSTR178. Genetic Techniques to Target and/or Manipulate Cells

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR178.11/VV14

Topic: I.01. Molecular, Biochemical, and Genetic Techniques

Support: Gatsby Charitable Foundation (562980)
Wellcome Trust (562763)

Title: Functional gene delivery using a novel Blood-Brain-Barrier-crossing AAV capsid in rats

Authors: *I. TOKSÖZ[1], *I. TOKSÖZ[1], T. K. AMAEE[1], D. RAZAGHI[1], V. PLATTNER[1], M. CHUAPOCO[2], X. CHEN[2], V. GRADINARU[3], A. AKRAMI[1];
[1]Sainsbury Wellcome Ctr., Univ. Col. London, London, United Kingdom; [3]Biol. and Biol. Engin., [2]Caltech, Pasadena, CA

Abstract: Transgenic rodent models are essential for neuroscientific research. The choice of exact species in a study will depend on the research question and behavioral demands, as well as amenability to experimental manipulations. Rats, equipped with a large behavioral repertoire

with varying complexity, can serve as a highly suitable animal model, especially for studies that require more advanced cognitive or behavioral tasks. Some transgenic rat strains are already commercially available; however, engineering and breeding such transgenic lines are cost and time intensive, highlighting the need for developing genetic tools targeting rats as an alternative to transgenic animals. Locally injectable Adeno-associated viruses (AAVs) are commonly used vehicles for gene delivery with modifiable features and numerous advantages, including target specificity, safety, and transduction efficiency. However, stereotaxic injection of AAVs is considered a highly invasive method, and there are several potential risks associated with it, including the risk of infection, brain damage, and the need for prolonged general anaesthesia. Furthermore, high variability between individual injections and among experimenters is inevitable, and virus spread cannot be fully controlled by the experimenter. To overcome these issues, developing blood-brain barrier (BBB) crossing and cell-type specific AAVs is particularly important as they can enter the brain tissue upon minimally invasive intravenous delivery (e.g. tail injections) and with more precisely controlled sparseness of transduced neurons. One example of such an AAV is CAP-Mac, previously shown to be highly neuron-specific and to have high transduction efficiency in the rat brain when packaged with fluorescent reporters (Chuapoco et al., 2023). In this study, we focused on the functional testing of CAP-Mac in optogenetic and calcium imaging settings. Acute and chronic extracellular recordings during light stimulations were used to functionally validate successful transduction of neurons with channelrhodopsins. Using a head-mounted widefield macroscope, cScope (Scott et al., 2018), fluorescence emitted from the genetically encoded calcium indicator, GCaMP6F, was imaged across the dorsal surface. AAV-delivered channelrhodopsin and GCaMP6F expressions were visualized and quantified using serial two-photon microscopy and immunohistochemistry methods. Our findings provide information on CAP-Mac's transduction efficiency and target preference in different conditions (sex, age, titer, cargo) and pave the way for using CAP-Mac in future behavioral studies in freely moving rats.

Disclosures: I. Toksöz: None. **I. Toksöz:** None. **T.K. Amaee:** None. **D. Razaghi:** None. **V. Plattner:** None. **M. Chuapoco:** None. **X. Chen:** None. **V. Gradinaru:** None. **A. Akrami:** None.

Poster

PSTR178. Genetic Techniques to Target and/or Manipulate Cells

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR178.12/VV15

Topic: I.01. Molecular, Biochemical, and Genetic Techniques

Title: Neuronal Transduction in Non-Human Primate Brains using the Human Synapsin and CAG Promoters

Authors: *H. B. CARROLL, A. GILES, A. MERCER, Y. LIU, O. DANOS, J. BRUDER, J. B. SMITH, B. HOLLIDGE;
REGENXBIO, ROCKVILLE, MD

Abstract: To develop adeno-associated virus (AAV) vector gene therapies for neurologic diseases, understanding the transduction efficiency of AAVs in the brain is paramount. Here, we compare the transduction efficiency and cell-type specificity of transgene expression for AAV9 with the CAG and human synapsin (hSyn) promoters following intraparenchymal injection to the brains of non-human primates (adult cynomolgus macaques). Prior to stereotaxic AAV infusion, the non-human primates were confirmed to be seronegative for AAV9. Bilateral cannula guides were aligned to the target infusion sites in the hippocampus of the left hemisphere and putamen in the right hemisphere. The trajectories and target depths were confirmed by MRI and gadolinium was co-injected with the AAV to visualize spread of the infusate. After three weeks, tissues were harvested, and we assessed the biodistribution of AAV9-CAG-GFP and AAV9-hSyn-GFP throughout the central nervous system and periphery (heart, skeletal muscle, liver, and spleen) as well as the seroconversion of the non-human primates. Immunohistochemistry was performed on coronal brain sections to determine the cell-type specificity of transgene expression. HALO® AI image analysis was used to determine the percentage of GFP-positive neurons in the hippocampus and putamen. At a dose of ~$1x10^{12}$ genome copies (gc) in the hippocampus and putamen, AAV9-hSyn-GFP was equally effective at transducing neurons as a corresponding ~$1x10^{11}$ gc dose of AAV9 with the ubiquitous CAG promoter without any gross toxicity. Biodistribution was measured using digital droplet PCR (ddPCR) and showed results that corroborated the HALO analysis, demonstrating that the higher dose of AAV9-hSyn-GFP was similar to the lower AAV9-CAG-GFP dose by transgene expression. As expected, the higher dose of AAV9-hSyn-GFP had a higher number of AAV genomes compared with the lower doses of AAV9-hSyn-GFP and AAV9-CAG-GFP. Similar trends were seen for the putamen. These findings demonstrate the utility of using the hSyn promoter in AAV vectors for selectively transducing neurons in the hippocampus and putamen in non-human primate studies.

Disclosures: **H.B. Carroll:** A. Employment/Salary (full or part-time); REGENXBIO, Inc. **A. Giles:** A. Employment/Salary (full or part-time); REGENXBIO. **A. Mercer:** A. Employment/Salary (full or part-time); REGENXBIO. **Y. Liu:** A. Employment/Salary (full or part-time); REGENXBIO. **O. Danos:** A. Employment/Salary (full or part-time); REGENXBIO. **J. Bruder:** A. Employment/Salary (full or part-time); REGENXBIO. **J.B. Smith:** A. Employment/Salary (full or part-time); REGENXBIO. **B. Hollidge:** A. Employment/Salary (full or part-time); REGENXBIO.

Poster

PSTR178. Genetic Techniques to Target and/or Manipulate Cells

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR178.13/VV16

Topic: I.01. Molecular, Biochemical, and Genetic Techniques

Support: LG202106-01-04

Title: Epigenetic editing therapy to treat Amyotrophic lateral sclerosis

Authors: *Y. SHEN, D. KONG, S. CHEN, C. ZHOU;
Ctr. for Excellence in Brain Sci. and Intelligence Technol., Shanghai, China

Abstract: Amyotrophic Lateral Sclerosis (ALS), colloquially known as Lou Gehrig's disease, is a devastating neurodegenerative disorder that progressively affects motor neurons, ultimately leading to patient fatality. Current treatments for ALS are exceedingly limited, with an average patient life expectancy of merely 2 to 5 years. In response to this, our research aims to explore new approaches by utilizing epigenetic editing technology to develop novel gene-editing therapies for ALS.
Epigenetic editing technology is an emerging gene regulatory method, its uniqueness lies in its ability to durably regulate the expression of target genes without altering the fundamental DNA sequence. Thus, this research aims to employ this technology to develop novel epigenetic editing tools and lipid nanoparticle delivery systems specifically targeting the nervous system.
Our research will involve the following aspects: First, we will develop durable and efficient epigenetic editing tools to regulate genes associated with ALS; Secondly, we will use lipid nanoparticle (LNP) delivery platforms to accurately deliver these editing tools to targeted brain regions to regulate the expression of target genes; Finally, we will explore the actual therapeutic effects of these epigenetic editing tools in ALS mouse models.
In summary, the goal of this research is to provide new theoretical and practical foundations for gene therapy of ALS. Meanwhile, we hope to provide powerful references for the treatment of other nervous system diseases by developing specific delivery tools.

Disclosures: **Y. Shen:** A. Employment/Salary (full or part-time); Center for Excellence in Brain Science and Intelligence Technology (Institute of Neuroscience). **D. Kong:** None. **S. Chen:** None. **C. Zhou:** A. Employment/Salary (full or part-time); Center for Excellence in Brain Science and Intelligence Technology (Institute of Neuroscience).

Poster

PSTR178. Genetic Techniques to Target and/or Manipulate Cells

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR178.14/VV17

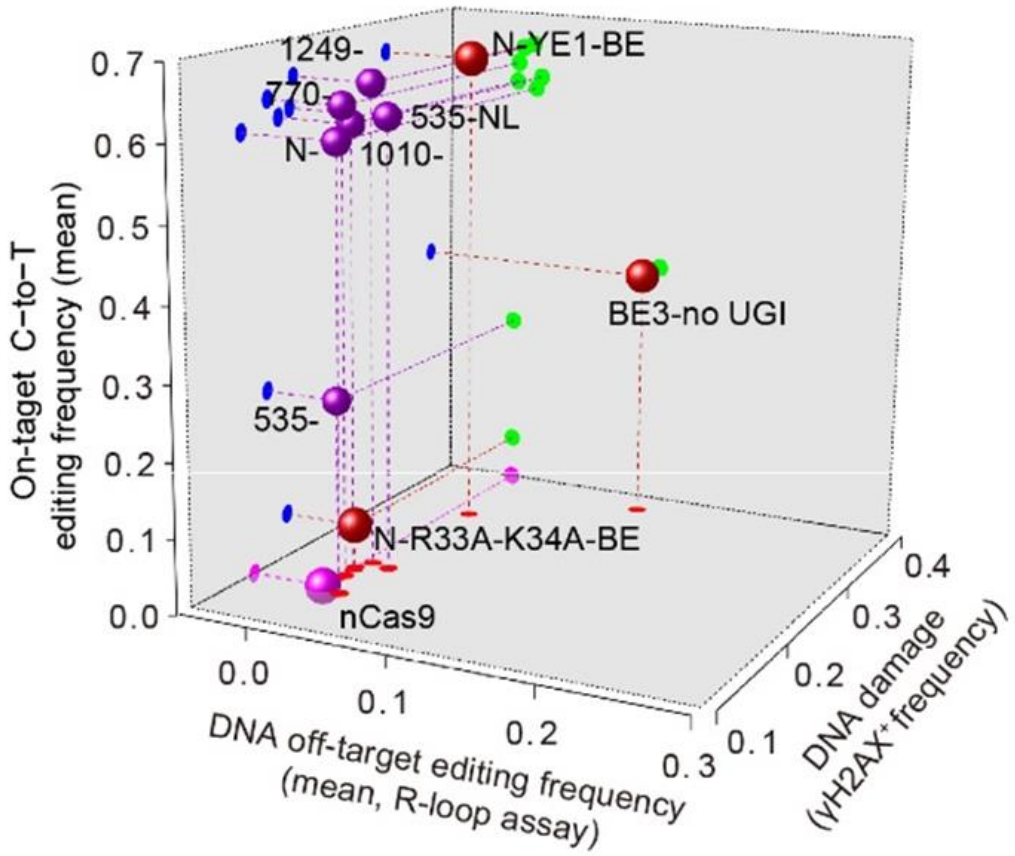
Topic: I.01. Molecular, Biochemical, and Genetic Techniques

Support: Grant No. LG-QS-202203-08
20JC1419500
20ZR1403100
2019YFA0111000
2020YFA0712403

Title: Engineering of cytosine base editors with DNA damage minimization and editing scope diversification

Authors: *S. ZHANG[1], L. SONG[2], B. YUAN[3], J. CHEN[1], J. CAO[2], J. QIU[1], Z. QIU[4,3,5,6], J. CHEN[2,7,8], X.-M. ZHAO[2,7,8], T.-L. CHENG[1];
[1]Inst. for Translational Brain Research, State Key Lab. of Med. Neurobiology, MOE Frontiers Ctr. for Brain Science, Inst. of Pediatrics, Natl. Children's Med. Center, Children's Hospital, Fudan Univ., Shanghai, China; [2]Inst. of Sci. and Technol. for Brain-inspired Intelligence, Key Lab. of Computat. Neurosci. and Brain-Inspired Intelligence, Fudan Univ., Shanghai, China; [3]Inst. of Neuroscience, State Key Lab. of Neuroscience, CAS Ctr. for excellence in Brain Sci. and Intelligence Technology, Chinese Acad. of Sci., Shanghai, China; [4]Songjiang Hospital, Songjiang Institute, Shanghai Jiao Tong Univ. Sch. of Med., Shanghai, China; [5]Clin. Neurosci. Center, Ruijin Hospital, Shanghai Jiao Tong Univ. Sch. of Med., Shanghai, China; [6]Natl. Clin. Res. Ctr. for Aging and Medicine, Huashan Hospital, Fudan Univ., Shanghai, China; [7]State Key Lab. of Med. Neurobiology, Inst. of Brain Science, Fudan Univ., Shanghai, China; [8]MOE Key Lab. of Computat. Neurosci. and Brain-Inspired Intelligence, and MOE Frontiers Ctr. for Brain Sci., Shanghai, China

Abstract: Cytosine base editors (CBEs), which enable precise C-to-T substitutions, have been restricted by potential safety risks, including DNA off-target edits, RNA off-target edits and additional genotoxicity such as DNA damages induced by double-strand breaks (DSBs), which can bring about limits to their applications in gene therapy for neurological diseases. Though DNA and RNA off-target edits have been ameliorated via various strategies, evaluation and minimization of DSB-associated DNA damage risks for most CBEs remain to be resolved. Here we demonstrate that YE1, an engineered CBE variant with minimized DNA and RNA off-target edits, could induce prominent DSB-associated DNA damage risks, manifested as γH2AX accumulation in human cells. We then perform deaminase engineering for two deaminases lamprey LjCDA1 and human APOBEC3A, and generate divergent CBE variants with eliminated DSB-associated DNA damage risks, in addition to minimized DNA/RNA off-target edits. Furthermore, the editing scopes and sequence preferences of APOBEC3A-derived CBEs could be further diversified by internal fusion strategy. We show that newly designed safer CBEs are able to introduce genetic mutations efficiently via adeno-associated virus delivery in the brain in vivo with few DNA off-target edits. Taken together, this study provides updated evaluation platform for DSB-associated DNA damage risks of CBEs and further generates a series of safer toolkits with diversified editing signatures to expand their applications in gene therapy for neurological diseases.



Disclosures: **S. Zhang:** None. **L. Song:** None. **B. Yuan:** None. **J. Chen:** None. **J. Cao:** None. **J. Qiu:** None. **Z. Qiu:** None. **J. Chen:** None. **X. Zhao:** None. **T. Cheng:** None.

Poster

PSTR178. Genetic Techniques to Target and/or Manipulate Cells

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR178.15/VV18

Topic: I.01. Molecular, Biochemical, and Genetic Techniques

Support: HUIDAGENE Therapeutics Inc.
HUIEDIT Therapeutics Inc.

Title: Development of miniature genome- and base-editing tools using an engineered IscB

Authors: ***D. HAN**[1,2], Q. XIAO[1,3,4], Y. WANG[1,2], H. ZHANG[3,4], X. DONG[3], G. LI[3], X. KONG[3,4], Y. YUAN[3], L. SHI[3], H. YANG[1,4,3], Y. ZHOU[3,4];
[1]Ctr. for Excellence in Brain Sci. and Intelligence Technol. (Institute of Neuroscience), Chinese Acad. of Sci., Shanghai, China; [2]Col. of Life Sciences, Univ. of Chinese Acad. of Sci., Beijing,

China; [3]HUIDAGENE Therapeut. Inc., Shanghai, China; [4]HUIEDIT Therapeut. Inc., Shanghai, China

Abstract: IscB, the likely ancestor of Cas9, has been characterized as a programmable RNA-guided endonuclease. It shares RuvC and HNH endonuclease domains with Cas9 but has only ~500 residues. Thus, IscB is a promising enzyme for developing effective miniature genome- and base-editing tools that are suitable for in vivo delivery. OgeuIscB has been reported to exhibited editing activity in HEK293FT cells. However, the efficiency is so poor for further application.In this study, we used a GFxxFP reporter system to screen IscB variants with higher efficiency in human cells. To improve IscB efficiency, We performed stem loop truncation and G-C base pairs substitution to optimize ωRNA structure. We also substituted residues of IscB with arginine to generate enhanced IscB protein. The enhanced IscB system (enIscB) combined optimized ωRNA and engineered protein, showing significantly improved editing efficiency compared with wild type OgeuIscB at 23 endogenous loci in HEK293T cells (35.934±27.354 verses 9.449±9.221, n = 3). By fusing enIscB with T5 exonuclease (T5E), we developed a enIscB-T5E system that exhibited comparable editing efficiency with SpG Cas9 at 23 TAM (NWRRNA)/PAM (NGN)-matched sites (60.61 ± 27.43 versus 53.25 ± 30.92%, n=3). Consistent with previous study, enIscB-T5E showed lower translocation rates compared with both enIscB and SpG Cas9. To engineer enIscB for miniature base editors, D61A (corresponding to D10A of SpCas9) was introduced to generate enIscB nickase. Next, we fused enIscB^{D61A} with TadA8e^{V106W} to develop several A-to-G DNA base editors with different architectures. The TadA8e^{V106W}-enIscB^{D61A}-TadA8e^{V106W} achieved the highest activity up to 52.37% at VEGFA locus and 60.06% at EMX1 locus in HEK293T cells respectively (n = 3), termed as miABE. Additionally, fusing enIscB with human APOBEC3A^{W104A} and uracil glycosylase inhibitor generated a miniature cytosine base editor (miCBE), which exhibited comparable C-to-T editing efficiency with SpG-CBE at 9 endogenous sites in HEK293T cells, which is 62.86 ± 47.13 versus 65.81 ± 65.81%. The specificity of miABE and miCBE were evaluated by Off-target analysis with in silico prediction and R-loop assay, in which similar levels of off-target events were detected compare with SpG-ABE and SpG-CBE respectively. These results indicated that enIscB system yields miniature and versatile tools for both genome editing and base editing in mammalian cells, which exhibit tremendous potential for basic research and biomedical applications.

Disclosures: **D. Han:** E. Ownership Interest (stock, stock options, royalty, receipt of intellectual property rights/patent holder, excluding diversified mutual funds); has filed patent applications related to this work through HUIDAGENE and CAS. **Q. Xiao:** E. Ownership Interest (stock, stock options, royalty, receipt of intellectual property rights/patent holder, excluding diversified mutual funds); has filed patent applications related to this work through HUIDAGENE and CAS.. **Y. wang:** None. **H. Zhang:** None. **X. Dong:** None. **G. Li:** None. **X. Kong:** None. **Y. Yuan:** None. **L. Shi:** Other; cofounder of HUIDAGENE Therapeutics. **H. Yang:** Other; cofounder of HUIDAGENE Therapeutics. and HUIEDIT Therapeutics. **Y. Zhou:** E. Ownership Interest (stock, stock options, royalty, receipt of intellectual property rights/patent holder, excluding diversified mutual funds); has filed patent applications related to this work through HUIDAGENE and CAS.. Other; cofounder of HUIEDIT Therapeutics.

Poster

PSTR178. Genetic Techniques to Target and/or Manipulate Cells

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR178.16/VV19

Topic: I.01. Molecular, Biochemical, and Genetic Techniques

Support: NIH Grant R01NS117585
NIH Grant R01NS123154

Title: Optimizing Prime Editing in human iPSCs for generating and correcting SCN2A genetic variations related to epilepsy and autism

Authors: *K. WETTSCHURACK[1], M. HALURKAR[1], Y. YANG[1], W. SKARNES[2]; [1]Medicinal Chem. and Mol. Pharmacol., Purdue Univ., West Lafayette, IN; [2]The Jackson Lab. For Genomic Med., Farmington, CT

Abstract: The transformative CRISPR-based technologies greatly facilitate the arrival of the genetic medicine era. Unlike traditional CRISPR, which generates a double-strand DNA break for genome editing, newly developed prime editing (PE) only creates a nick (single-strand DNA break). Thus, PE is considered to have an enhanced safety profile and holds enormous potential for precision genome editing to eventually correct disease-causing genetic variants in patients. Human induced pluripotent stem cells (hiPSCs) have emerged as a preferred system for disease modeling and testing for precision genetic interventions in cells with human genetic backgrounds. Although PE can perform genome editing in hiPSCs, the efficiency is far from ideal. Indeed, using PE to create or correct disease-causing genetic variants related to neurological disorders in hiPSCs-based models is still in its infancy. Here, we developed a fluorescent assay to monitor prime editing efficiency in hiPSCs. We nucleofected PE reagents that introduce the H67Y (C->T) change in iPSCs expressing blue fluorescent protein (BFP), converting BFP into green fluorescent protein (GFP). Edited vs unedited iPSCs can be easily quantified by flow cytometry. Following the co-delivery of epegRNA and plasmid-based prime editors, we demonstrated successful conversion of BFP to GFP. PE efficiency was improved 10-fold with the addition of nicking gRNA (ngRNA) from 4% to 40% of cells. We plan to use the BFP->GFP assay to test new reagents and accessory factors that may improve the efficiency of PE in iPSCs to obtain an optimal editing condition. We will then apply our optimized conditions to edit single nucleotide variants to create or correct disease-causing mutations in the voltage-gated sodium channel Nav1.2 (encoded by the SCN2A gene), which has recently been identified as the leading cause of monogenic autism and epilepsy. In conclusion, we demonstrated the successful use of prime editing in human iPSCs and provided a valuable assay for researchers to optimize the efficiency of prime editing and make precision modifications in iPSCs.

Disclosures: K. Wettschurack: None. **M. Halurkar:** None. **Y. Yang:** None. **W. Skarnes:** None.

Poster

PSTR178. Genetic Techniques to Target and/or Manipulate Cells

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR178.17/VV20

Topic: I.01. Molecular, Biochemical, and Genetic Techniques

Title: Functional genomic CRISPR knockout screens in iPSC-derived ioMicroglia

Authors: H. CEYLAN, N. PAPAI, K. ARAT, C. SCHMIDT, ***A. BYRNE**, S. SALIC, T. BURCKSTUMMER;
bit.bio discovery, Vienna, Austria

Abstract: Huriye Ceylan[1], Nora Papai[1], Kemal Arat[1], Clara Schmidt[1], Nikola Vinko[1], Tatyana Perlova[1], Ann Byrne[1], Sejla Salic[1], Tilmann Bürckstümmer[1] [1] bit.bio discovery, Vienna, Austria Microglia play an integral role in neuroinflammation, phagocytosis, synaptic regulation, and neurotoxicity modulation. As many of these processes become dysregulated in disease, microglia have emerged as promising therapeutic targets for a diverse range of neurological disorders. Leveraging induced pluripotent stem cell (iPSC) derived ioMicroglia, which provide a physiologically relevant model, we introduce bit.bio discovery's innovative CRISPR screening platform. Functional genomic screening using CRISPR/Cas9 is a powerful tool in deciphering gene function, identifying disease mechanisms, and uncovering novel therapeutic targets. Here, we apply our workflow in iPSC-derived ioMicroglia to create screenable Cas9 expressing cells. We demonstrate that expression of Cas9 has no effect on the transcriptomic profile, differentiation potential or cytokine production of lipopolysaccharide (LPS) stimulated ioMicroglia. We provide proof-of-concept that functional genomic screening can be performed at both iPSC stage prior to forward programming and following forward programming in ioMicroglia. Utilising these cells, we plan to perform a pooled CRISPR knockout screen of 200 genes involved in the modulation of innate immune signaling and LPS response. Readouts will include both whole transcriptome and targeted single-cell RNA sequencing, FACS analysis of surface marker expression and cytokine production following LPS stimulation. Our results highlight the power of unbiased genetic screens in iPSC-derived cells and provides a robust platform for systematic interrogation of normal and diseased cell states for disease modelling and target discovery.

Disclosures: **H. Ceylan:** None. **N. Papai:** None. **K. Arat:** None. **C. Schmidt:** None. **A. Byrne:** None. **S. Salic:** None. **T. Burckstummer:** None.

Poster

PSTR178. Genetic Techniques to Target and/or Manipulate Cells

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR178.18/VV21

Topic: I.01. Molecular, Biochemical, and Genetic Techniques

Support: NIH Grant R01 NS130046

Title: A tool for single-vector CRISPR/Cas9 editing and transgene expression

Authors: *J. MOFFA, I. BLAND, V. KALYANARAMAN, J. TOOLEY, M. CREED, B. COPITS;
Anesthesiol., Washington Univ. in St. Louis, St. Louis, MO

Abstract: Neuronal gene manipulation is a crucial tool for advancing our understanding of the nervous system. However, current techniques can be costly, may lack cell-type specificity, and allow limited manipulation of edited neurons. To overcome these limitations, we have developed an alternative gene editing strategy using a single AAV vector and mouse lines that express Cre-dependent Cas9 to achieve efficient cell-type specific editing across the nervous system. Expressing Cre-dependent Cas9 in specific cell types in transgenic mouse lines frees up space to package the guide RNAs for gene editing with a Cre-dependent marker gene in a single virus. This permits target gene editing and visualization and/or manipulation of edited cells using a single viral injection. These vectors express Cre-dependent fluorophores for tracing (mCherry, synaptophysin-GFP), targeted recordings (NLS-Ruby), calcium imaging (GCaMP8f), and optogenetic experiments (ChRonos).
We tested our strategy in multiple brain regions and cell types, including GABAergic neurons in the nucleus acumbens (NAc), glutamatergic neurons projecting from the ventral pallidum (VP) to the lateral habenula (LHb), dopaminergic neurons in the ventral tegmental area (VTA), and parvalbumin (PV)-positive proprioceptive neurons in the periphery. In GABAergic NAc neurons and glutamatergic VP::LHb neurons, we used optogenetic stimulation in acute slices to demonstrate efficient editing of *Vgat*, which significantly reduced evoked inhibitory postsynaptic currents. In dopaminergic VTA neurons, we expressed the fluorescent Ca^{2+} sensor GCaMP8f and showed that knockdown of *Grin1*, an essential NMDA receptor subunit, significantly reduced the amplitude of Ca^{2+} transients in response to both a tone cue and reward in awake, behaving mice. Finally, we combined our approach with newly-developed peripheral AAV serotypes by expressing mCherry in peripheral proprioceptive neurons of p1 mice and editing the *Dicer1* gene. This manipulation resulted in retraction of centrally-projecting axons from the ventral horn of the spinal cord, demonstrating the utility of our approach for both circuit-tracing and PNS applications.
These data demonstrate the efficacy of our single-vector approach for gene editing and transgene expression. This method allows researchers to combine CRISPR/Cas9 gene editing with tools to visualize neurons, manipulate their activity, or record their responses to stimuli, improving our ability to study novel genes of unknown significance and advance our understanding of the nervous system.

Disclosures: J. Moffa: None. **I. Bland:** None. **V. Kalyanaraman:** None. **J. Tooley:** None. **M. Creed:** None. **B. Copits:** None.

Poster

PSTR178. Genetic Techniques to Target and/or Manipulate Cells

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR178.19/VV22

Topic: I.01. Molecular, Biochemical, and Genetic Techniques

Support: NIH grant DP2 MH122398

Title: Tag You're It!: Comparative analysis of knockin tags for endogenous protein visualization using Cas9-RC in the mouse brain

Authors: *A. J. ROMANOWSKI[1], R. R. RICHARDSON[2], A. POULOPOULOS[3];
[1]Univ. of Maryland Sch. of Med., Baltimore, MD; [2]NIH, Baltimore, MD; [3]Univ. of Maryland SoM, Univ. Maryland, Baltimore, MD

Abstract: Advancements in CRISPR technologies have enabled the creation of high-performance in vivo editing agents that can introduce tags onto genes of wildtype animals without the need for transgenesis and animal lines. By significantly enhancing knockin precision, efficiency, and delivery, this technology has opened up new avenues for studying endogenous proteins in the central nervous system (CNS) of mice. However, the current challenge lies in selecting the optimal knockin payload that ensures the highest signal-to-noise ratio in brain tissue for the visualization of endogenous proteins. There are three primary categories of tags that are commonly employed: 1) small epitope tags like HA, Myc, and ALFA, 2) large fluorescent proteins such as eGFP and mScarlet, and 3) self-labeling protein tags like HALO and fluorogen activating peptides (FAP). To evaluate the effectiveness of tags from each group, we conducted a comparative analysis on two distinct loci with different expression patterns in mouse cortex. For our high-expressing locus, we selected β-Actin (ActB), which exhibits ubiquitous high expression in neurons and glia. For our low-expressing locus, we chose Neuronal Growth Regulator 1 (Negr1), a GPI-anchored membrane protein that displays variable expression levels during development and into adulthood in cortex. To achieve precise knockin at each target locus, we used in utero electroporation with Cas9-RC, a Cas9-based fusion protein that increases knockin efficiency and precision (Richardson et al. 2023). Following the introduction of the tag payloads, we compared the signal produced by each tag against the background to determine the tag with the optimal signal-to-noise ratio in tissue slices for high- and low-expressing loci.

Disclosures: A.J. Romanowski: None. **R.R. Richardson:** None. **A. Poulopoulos:** None.

Poster

PSTR178. Genetic Techniques to Target and/or Manipulate Cells

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR178.20/VV23

Topic: I.01. Molecular, Biochemical, and Genetic Techniques

Support: NIH DP2MH122398
NIH R01DA039062

Title: All genes are not equal: "hub" transcriptional regulators are unusually robust to CRISPR activation

Authors: *A. E. MARQUARDT[1,2], A. J. ROMANOWSKI[1,2], B. ALTAS[2], M. CONTRERAS[3], M. M. MCCARTHY[1,2], A. POULOPOULOS[1,2];
[1]Univ. of Maryland Med. - Inst. for Neurosci. Discovery, Baltimore, MD; [2]Dept. of Pharmacol., [3]Dept. of Physiol., Univ. of Maryland Sch. of Med., Baltimore, MD

Abstract: The advent of CRISPR-Cas9 technology has revolutionized many areas of neuroscience research via the creation of novel tools to manipulate expression of endogenous genes. One such tool, CRISPR activation (CRISPRa), has harnessed the power of the CRISPR-Cas9 system to allow for gene-specific transcriptional activation. We recently utilized this system with the goal of exogenously upregulating candidate “hub genes” - highly interconnected genes within a transcriptional module thus predicted to control expression of other module genes - identified using weighted gene co-expression network analysis (WGCNA) following an RNA-sequencing study of genes associated with social play. We designed CRISPR strategies targeting 6 candidate transcriptional hub genes - *Spen*, *Kmt2d*, *Rere*, *Ubr4*, *Cabin1*, and *Dip2b* - as well as *Cyp19a1*, an unrelated “control” gene not identified by our WGCNA. We subcloned 3-6 CRISPRa single guide RNAs (sgRNAs) per gene into a *Sa*Cas9-compatible backbone. Alongside a d*Sa*Cas9-VPRmini plasmid successfully used by others, we nucleofected these CRISPRa agents individually into primary rat cortical neurons, then quantified target gene expression using qPCR at 7 days *in vitro* (DIV). Using these methods, multiple *Cyp19a1*-targeting sgRNAs individually elicited a potent (~10x) upregulation of the *Cyp19a1* control gene, and in combination displayed dose-dependent upregulation. Surprisingly, out of the twenty-two sgRNAs targeting the six transcriptional hub genes, none resulted in appreciable gene activation. We assessed each of the sgRNAs individually, as well as multiple combinations thereof, yet none successfully upregulated any of the six candidate hub genes, despite the fact that this same system was successful for the control gene *Cyp19a1*. Thus, we postulate that endogenous regulatory elements may more tightly limit manipulation of gene expression levels for hub transcriptional regulators, making them less amenable to CRISPRa. This hypothesis is supported by data from ENCODE indicating a much more complicated landscape of candidate cis-regulatory elements (cCREs) for the six hub genes as compared to *Cyp19a1*. To test this, we will clone the sgRNA target regions of each hub gene into a common transcriptional reporter system to quantify upregulation. We predict that at least some of the CRISPRa agents that failed in the endogenous system will be successful in the absence of endogenous regulatory constraints, indicating that genes key to fate commitment, like our identified hub genes, may have evolved tight endogenous titration mechanisms limiting variance in gene dosage.

Disclosures: **A.E. Marquardt:** None. **A.J. Romanowski:** None. **B. Altas:** None. **M. Contreras:** None. **M.M. McCarthy:** None. **A. Poulopoulos:** None.

Poster

PSTR178. Genetic Techniques to Target and/or Manipulate Cells

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR178.21/VV24

Topic: I.01. Molecular, Biochemical, and Genetic Techniques

Support: NIH Grant DP2 MH122398

Title: In vivo CRISPR agents against glioma: development and delivery

Authors: *R. WHITTEN[1], A. ROMANOWSKI[2], C. BRANDENBURG[3], A. P. MALLA[4], G. WOODWORTH[5], P. ANASTASIADIS[5], A. POULOPOULOS[6];
[1]Neurosci., [2]Univ. of Maryland Baltimore, Baltimore, MD; [3]Pharmacol., Univ. of Maryland Sch. of Med. Program In Neurosci., Baltimore, MD; [4]Mol. Med., [5]Neurosurg., Univ. of Maryland, Baltimore, MD; [6]Pharmacol., Univ. Maryland, Baltimore, MD

Abstract: In vivo CRISPR agents against glioma: development and delivery
Ro Whitten[1], Andrea Romanowski[1], Cheryl Brandenburg[1], Adarsha Malla[2,3], Graeme Woodworth[2,3], Pavlos Anastasiadis[2,3], Alexandros Poulopoulos[1]
[1]Department of Pharmacology and UM-MIND, University of Maryland School of Medicine, Baltimore, MD, USA [2]Department of Neurosurgery and UM-MIND, University of Maryland School of Medicine, Baltimore, MD, USA [3]University of Maryland Marlene and Stewart Greenebaum Comprehensive Cancer Center, Baltimore, MD, USA
Dual landmark advances in tumor microenvironment research in the brain and CRISPR neural somatic genome editing technologies have created a unique opportunity for targeting brain tumors with genome therapeutics. Unexpectedly, a protein released from brain cells by synaptic activity, Neuroligin-3 (NLGN3), was identified as a necessary component of the tumor microenvironment for the growth and spread of aggressive forms of glioma, including glioblastoma (Venkatesh et al. *Cell*, 2015). The effect is that glioblastoma xenografts fail to grow in Nlgn3 knockout mice (Venkatesh et al. *Nature* 2017). Through our investigations into the synaptic functions of Nlgn3, we have developed antibodies and CRISPR agents able to target Nlgn3 acutely in wild-type mice and rats. This study pursues the development of the technological capabilities to deliver CRISPR agents to the adult mouse brain non-invasively using focused ultrasound. Using this development, we begin to pursue neural somatic genome editing therapeutics for brain pathologies.

Disclosures: R. Whitten: None. **A. Romanowski:** None. **C. Brandenburg:** None. **A.P. Malla:** None. **G. Woodworth:** None. **P. Anastasiadis:** None. **A. Poulopoulos:** None.

Poster

PSTR178. Genetic Techniques to Target and/or Manipulate Cells

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR178.22/VV25

Topic: I.01. Molecular, Biochemical, and Genetic Techniques

Support: E. Malcolm Field and Gary Leo Dunbar Endowed Chair in Neuroscience at Central Michigan University, John G Kulhavi Professorship, Neuroscience Program, College of Medicine
Office of Research and Graduate Studies at Central Michigan University.

Title: Antineoplastic Effect of AVIL Knockout in U87 Human Glioblastoma Cells using CRISPR/Cas9

Authors: *A. POUDEL, J. E. SMITH, C. N. NOE, B. SRINAGESHWAR, J. L. BAKKE, G. L. DUNBAR, J. ROSSIGNOL;
Central Michigan Univ., Mt. Pleasant, MI

Abstract: Glioblastoma multiforme (GB) is a grade IV astrocytoma, the most common and malignant adult central nervous system tumor. Current standard treatment for GB includes cytoreduction surgeries by surgical resection of the tumor, radiation therapy, and temozolomide (TMZ) therapy, which extends patient life expectancy by 12-15 months with 95% mortality within five years. TMZ is a chemotherapy which accounts for most of the side effects of a chemotherapy drug including hair loss, loss of fertility, weight loss, fatigue and more. In recent years, GB research has focused more on gene therapies. One gene identified to have major role in contributing GB is *AVIL* which encodes protein Advillin (p92). P92 is a member of the gelsolin/villin family, an acting binding protein. *AVIL* is overexpressed in almost every GB cell and is essential for proliferation and migration of GBs. In patients with GB, higher expression of *AVIL* correlates with worse outcomes. In this study, we knocked-out *AVIL* in U87 human glioblastoma cells, *in vitro* using CRISPR/Cas9 gene editing tool and investigated its effects on downstream proteins. (1) Foxm1which is a key regulator of mitosis and its overexpression of Foxm1 promote cell cycle progression; and (2) tumor suppressor protein p53. We used Lipofectamine CRISPRMAX to transfect U87 human glioblastoma cells with *AVIL*-CRISPR/Cas9. Transfected U87 cells were analyzed for knockout via Sanger sequencing and for protein expression using Western blot and immunocytochemistry (ICC). Sanger sequencing of transfected U87 showed 55% knockout after 3 days of transfection. We used HEKT293 cells as controls, which showed 22% knockout under the same conditions. Western blot showed reduced expression of p92 protein in U87, as well as in HEKT293 cells, confirming the gene knockout and reduced expression of downstream protein Foxm1 in both U87 and HEKT293 cells. ICC results revealed increased expression of p53 protein expression. The results suggested that *AVIL* knockouts were able to reduce the Advillin expression, which reduced the activity of Foxm1 and increased the activity of tumor suppressor protein p53. This preliminary study shows that CRISPR/Cas9-mediated *AVIL* knockout as a novel and potential antitumor treatment option for GB.

Disclosures: A. Poudel: None. **J.E. Smith:** None. **C.N. Noe:** None. **B. Srinageshwar:** None. **J.L. Bakke:** None. **G.L. Dunbar:** None. **J. Rossignol:** None.

Poster

PSTR178. Genetic Techniques to Target and/or Manipulate Cells

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR178.23/VV26

Topic: I.01. Molecular, Biochemical, and Genetic Techniques

Support: NIH Grant 1R03NS123733

Title: Harnessing ICyCam for Unraveling Glioblastoma Complexity: An Advanced Approach for Single-Cell Tracking

Authors: J. WANG, P. WALCZAK, ***Y. LIANG**;
Univ. of Maryland Sch. of Med., Baltimore, MD

Abstract: Glioblastoma (GBM) heterogeneity significantly contributes to tumor recurrence and therapy resistance. This study introduces an innovative application of intravital imaging and labeling methodologies to elucidate GBM heterogeneity, featuring a long-term intravital single-cell tracking platform, 2-photon microscopy (2pLIST). Our exploration centers on monitoring three key parameters, cell cycle, calcium, and identity, simultaneously utilizing genetically encoded indicators. The Fluorescent Ubiquitination-based Cell Cycle Indicator (FUCCI) collaborates with color-compatible green or red calcium indicators. In the quest for effective cell identity tracking, we initially trialed two strategies for giving cells a unique identity for tracking: RGB labeling and photoactivatable fluorophores, with the latter proving superior due to RGB's inefficacy over proliferative periods. Consequently, we adopted photoactivatable GFP (PAGFP) for durable GBM cell tracking. To achieve spectral and spatial cell labeling multiplexing, we combined BFP-mCherry based FUCCI (located in the nucleus), jRGECO1a (in cytosol), and PAGFP (in cytosol), termed as ICyCam (identity, cell cycle, and calcium). The distinct cellular localizations of jRGECO and FUCCI facilitated their differentiation. Moreover, jRGECO and PAGFP were distinguishable by their spectra. We characterized the lentiviral construct encoding ICyCam in culture and validated its in vivo efficacy for single-cell tracking of human GBM in immunocompetent mice. ICyCam has emerged as an invaluable labeling tool for 2pLIST, providing intricate and dynamic insights into glioblastoma behavior in live animal models. It holds the potential to enhance understanding of tumor cell heterogeneity, pathogenesis, and foster advancements in in vivo drug screening.

Disclosures: J. Wang: None. **P. Walczak:** None. **Y. Liang:** None.

Poster

PSTR178. Genetic Techniques to Target and/or Manipulate Cells

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR178.24/VV27

Topic: I.01. Molecular, Biochemical, and Genetic Techniques

Title: A rapid method for identification of APOE alleles ε2, ε3, and ε4 via electrochemiluminescence

Authors: *F. KUNG, N. ENCARDES, T. J. BREAK, S. B. HARKINS, J. DEBAD, J. N. WOHLSTADTER;
Meso Scale Discovery, Rockville, MD

Abstract: Apolipoprotein E, a lipoprotein encoded by human gene APOE, is the principal cholesterol carrier in the brain. Three alleles (ε2, ε3, and ε4) have been identified as risk factors for Alzheimer's disease and are differentiated by single nucleotide polymorphisms (SNPs) at two sites in the gene. A multiplex assay was developed to quickly distinguish between the three alleles as an alternative to sequencing, qPCR, or traditional oligonucleotide ligation assay (OLA) methods.

A single amplicon that spans both APOE mutation sites was generated using a fast PCR protocol (runtime ≈50 minutes). For each target, two probes that differ only at the polymorphic base were tagged with capture sequences specific for spots on an N-PLEX plate and paired with a probe that contained a 5' phosphate for ligation and a 3' biotin for detection. These probes were combined for the OLA (initial denaturation and 30 cycles of ligation: total ≈1.5 hours) or a single ligation (initial denaturation and static temperature 15 minutes). The product was transferred to 96-well, 10-spot N-PLEX plates for hybridization of the ligated probes to their corresponding plate-bound oligonucleotides. SULFO-TAG labeled streptavidin was then bound to biotin on ligated probes, and plates were read on an MSD instrument for electrochemiluminescence detection (total optimized assay time ≈3.5 hours).

The OLA and single-ligation methods were able to clearly discriminate single-base changes at both SNP sites in 79 extracted blood samples. Allele frequencies were 6.3% (ε2), 77.5% (ε3), and 16.3% (ε4), similar to previously reported U.S. data. Additionally, the ε4/ε4 genotype, shown to be a significant risk factor in the development of late-onset Alzheimer's disease, was 1.4%. The genotyping results were confirmed by Sanger sequencing. This optimized protocol shortened total assay time from ≈5.5 hours to ≈3.5 hours, while still allowing for clear base discrimination at both SNP sites.

This report highlights a novel method to quickly identify APOE genotypes. The assay improves sample throughput, making this a viable alternative to sequencing and other methods. Lastly, this method can be easily adapted to target SNPs or insertions/deletions that are identified as risk factors for other diseases.

Disclosures: **F. Kung:** A. Employment/Salary (full or part-time); Meso Scale Diagnostics, LLC. **N. Encardes:** A. Employment/Salary (full or part-time); Meso Scale Diagnostics, LLC. **T.J. Break:** A. Employment/Salary (full or part-time); Meso Scale Diagnostics, LLC. **S.B. Harkins:** A. Employment/Salary (full or part-time); Meso Scale Diagnostics, LLC. **J. Debad:** A. Employment/Salary (full or part-time); Meso Scale Diagnostics, LLC. **J.N. Wohlstadter:** A. Employment/Salary (full or part-time); Meso Scale Diagnostics, LLC.

Poster

PSTR178. Genetic Techniques to Target and/or Manipulate Cells

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR178.25/VV28

Topic: I.01. Molecular, Biochemical, and Genetic Techniques

Support: NIH R35GM140854
NIH R01HL153916-01A1
University of Virginia Double Hoo Award
University of Virginia EXPAND Fellowship
University of Virginia Presidential Fellowship

Title: Development of Humanized Glp-1 Receptor Mouse to investigate the small molecule agonist mechanism of action

Authors: *E. GODSCHALL[1], A. BUYUKAKSAKAL[2], I. SAJONIA[3], A. SPANO[3], A. KEELER[3], J. CAMPBELL[3], C. DEPPMANN[3], A. D. GULER[4];
[1]Univ. of Virgina, Charlottesville, VA; [2]Neurosci. Undergraduate Program, [4]Biol., [3]Univ. of Virginia, Charlottesville, VA

Abstract: Western diets and lifestyles increase the rates of Type II Diabetes and obesity, necessitating efficacious identification of drug targets. Glucagon-like peptide-1 receptor (Glp-1R) agonists stimulate the release of insulin from the pancreas, decrease blood glucose, and induce satiety and weight loss. While the FDA-approved Glp-1R treatments are mainly injectable peptides, orally administered small molecule agonists will be coming to the market soon. Danuglipron, an orally-administered small molecule, requires a tryptophan residue in the amino acid position 33 of Glp-1R for its activity, which is present in the human Glp-1 receptor but replaced by serine in the mouse Glp-1R. To evaluate Danuglipron's mechanisms of action in the periphery and the brain of mice, using CRISPR-Cas9 technology, we introduced a single mutation in the mouse Glp-1R converting Ser33 to Trp33—functionally humanizing the receptor for Danuglipron action. We demonstrated that this mutated Glp-1R allele is heritable across multiple generations and that homozygous mice do not differ in terms of body weight, metabolism, or feeding compared to wild type mice. Additionally, Danuglipron administration blunts acute food intake while increasing glucose tolerance in these mice, validating the functionality of our novel Glp-1R mutated mouse model. Lastly, we investigated the capacity of Danuglipron to activate hypothalamic and hindbrain regions to better understand its functional access to the satiety and nausea-inducing circuits in the brain, revealing potential central mechanisms of action for this class of diabetes and weight-loss drugs.

Disclosures: E. Godschall: None. **A. Buyukaksakal:** None. **I. Sajonia:** None. **A. Spano:** None. **A. Keeler:** None. **J. Campbell:** None. **C. Deppmann:** None. **A.D. Guler:** None.

Poster

PSTR178. Genetic Techniques to Target and/or Manipulate Cells

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR178.26/VV29

Topic: I.01. Molecular, Biochemical, and Genetic Techniques

Support: F31AG081046
T32NS096050
AG079199
AG061175
5R01AG075820

Title: Developing a novel noradrenergic-specific proteomic mouse model

Authors: *A. KORUKONDA[1], C. C. RAMELOW[2], P. KUMAR[2], L. CHENG[2], C. LILES[1], S. RANGARAJU[2], D. WEINSHENKER[1];
[1]Human Genet., [2]Neurol., Emory Univ., Atlanta, GA

Abstract: The mammalian brain is defined by considerable cellular heterogeneity. Recent advances in profiling techniques allow the resolution of regional differences. The cell type-specific *in vivo* biotinylation of proteins (or CIBOP) utilizes the biotin ligase, TurboID, to label proximal native state proteins in the desired cell population. The brainstem noradrenergic locus coeruleus (LC) modulates several critical physiological and affective processes, and LC-Norepinephrine (NE) dysfunction has been implicated in various neuropsychiatric and neurodegenerative disorders. However, the mouse LC contains only ~1500 neurons per hemisphere, making it difficult to characterize biochemically in pathological conditions. Here, we have developed a novel transgenic strategy to confine expression of TurboID within LC-NE cells, allowing cell-type specific proteomic analysis for broader biological applications. *Rosa26*$^{TurboID/wt}$ mice were crossed to tyrosine hydroxylase (TH)-cre animals to enable cre recombination specifically in catecholaminergic TH-producing cells. Three-month old TH-cre/*Rosa26*$^{TurboID/wt}$ and littermate controls (n = 3/group) received biotin supplementation (37.5 mg/L) in drinking water for 2 weeks, following which brain tissue was processed for western blots and immunohistochemistry (IHC) to confirm biotinylation signal in the region of interest. Western blot lysates of whole brainstem tissue displayed robust endogenous biotinylation in the TH-cre/*Rosa26*$^{TurboID/wt}$ mice in comparison to control mice (TH-cre and *Rosa26*$^{TurboID/wt}$ groups). Immunoprecipitation with streptavidin beads enriched for biotinylated proteins in the brainstem lysates compared to littermate controls, which was confirmed with western blots and silver stains. Additionally, biotin labeling using a streptavidin-conjugated fluorophore was restricted to the cell bodies and dendrites of TH$^+$ cells in the LC. Converging data from the western blots and IHC indicate that the biotinylated proteins obtained from the brainstem dissection can solely be sourced from the LC. These data demonstrate that the transgenic TH-cre/*Rosa26*$^{TurboID/wt}$ mouse model can provide cell-type specific proteomic information from LC-NE neurons. The next steps involve characterizing and quantifying the biotinylated proteins from the LC via mass spectrometry. Future applications of this project include leveraging this model to identify proteomic LC responses to genetic and environmental challenges.

Disclosures: A. Korukonda: None. **C.C. Ramelow:** None. **P. Kumar:** None. **L. Cheng:** None. **C. Liles:** None. **S. Rangaraju:** None. **D. Weinshenker:** None.

Poster

PSTR178. Genetic Techniques to Target and/or Manipulate Cells

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR178.27/VV30

Topic: I.01. Molecular, Biochemical, and Genetic Techniques

Support: 1R01NS107428

Title: Alterations in Kelch Like Protein 13 (KLHL13) disrupt cell cycle regulation and cause developmental delay along with macrocephaly in humans and zebrafish

Authors: *T. AKHTER[1,2], A. GHAFFAR[3], A. SCHMIDT[4], J. O. MURPHY[6], M. B. PALOMARES[7], H. ENGELS[4], A. MERON[6], K. CREMER[4], E. MANGOLD[5], S. PETERS[5], Z. M. AHMED[1], S. RIAZUDDIN[8,2], S. RIAZUDDIN[1];
[1]Otolaryngology Head and Neck Surgery, Univ. of Maryland, Sch. of Med., Baltimore, MD; [2]Ctr. of Excellence in Mol. Biology, Univ. of the Punjab, Lahore, Pakistan; [3]Univ. of Maryland Sch. of Med., Baltimore, MD; [4]Inst. of Human Genet., [5]Univ. of Bonn, Bonn, Germany; [6]The Huntington's Dis. Ctr., Univ. of Pennsylvania, Philadelphia, PA; [7]INGEMM -Institute of Med. and Mol. Genet., La Paz Univ. Hosp., Madrid, Spain; [8]Jinnah Burn and Reconstructive Surgery Center, Allama Iqbal Med. College, Univ. of Hlth. Sci., Lahore, Pakistan

Abstract: Background Intellectual disability (ID) affects 1-3% of the world's population and monogenic causes are characterized by high genetic heterogeneity which challenges clinical and genetic diagnosis. *KLHL13* encodes Kelch Like Protein 13 which is a substrate adaptor for chromosomal passenger Aurora Kinase B (AURKB) in the E3-ubiquitin ligase complex. This complex is essential for the ubiquitination of AURKB to facilitate proper cell cycle regulation. **Approach:** We searched for the individuals carrying *KLHL13* variants with an international matchmaking arrangement initiated by GeneMatcher. We performed cell cycle synchronization and flow cytometry analyses to investigate the impact of ID-associated *KLHL13* variants on cell cycle regulation. Further, we knock down *klhl13* in a transgenic zebrafish line using a morpholino-based approach to determine its functional importance in early development. Finally, a dose-response of a selective pharmacological AURKB inhibitor AZD1152-HQPA was used to rescue phenotypic abnormalities of *KLHL13* loss of function, *in vitro* and *in vivo*. **Results:** Here, we report one frameshift c.1781-1782delTG and three missenses c.953C>T, c.179A>G, c.557G>A rare hemizygous variants in *KLHL13* causing mild to severe ID, developmental and speech delay, macrocephaly, unsteady gait and facial dysmorphism in seven individuals of four families from different ethnicities. *In vitro,* analyses of WT and variants overexpressing COS7 cells showed chromosomal missegregation at anaphase and formation of multinucleated cells at cytokinesis, supported by flow cytometric analysis revealed polyploidy at M/G1 phase leading to genomic instability. Furthermore, we observed that instead of at the central spindle AURKB was accumulated to chromosomal arms during anaphase in the variants which caused incomplete cytokinesis indicating AURKB as the main reason for abnormal cell cycle regulation. *klhl13* knockdown in zebrafish resulted in developmental delay, macrocephaly, facial dysmorphism, and neurobehavioral deficits comparable to human subjects. Human *KLHL13*^{WT} mRNA microinjections significantly rescued developmental and behavioral deficits in *klhl13* morphants (MO). However, the variants mRNAs were unable to rescue the observed MO phenotypes. Interestingly, AZD1152 treatment significantly restored genomic stability with AURKB localization in variants overexpressing heterologous cells and developmental deficits in MO.

Conclusion: These findings suggest that *KLHL13* has neurodevelopmental importance in humans and zebrafish, and inhibition of AURKB can be a potential treatment for individuals suffering from variants in *KLHL13*.

Disclosures: T. Akhter: None. **A. Ghaffar:** None. **A. Schmidt:** None. **J.O. Murphy:** None. **M.B. Palomares:** None. **H. Engels:** None. **A. Meron:** None. **K. Cremer:** None. **E. Mangold:** None. **S. Peters:** None. **Z.M. Ahmed:** None. **S. Riazuddin:** None. **S. Riazuddin:** None.

Poster

PSTR178. Genetic Techniques to Target and/or Manipulate Cells

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR178.28/VV31

Topic: I.01. Molecular, Biochemical, and Genetic Techniques

Support: CONACYT CF-2019-6390

Title: Genetic Relationship between Cortical Surface Area and Cortical Thickness in the Mexican Population

Authors: *I. ESPINOSA MÉNDEZ[1,2], T. V. ROMÁN LÓPEZ[1], D. RAMÍREZ GONZÁLEZ[1], I. C. SANCHÉZ MONCADA[1], X. DÍAZ TÉLLEZ[2,3], C. DOMÍNGUEZ FRAUSTO[1], V. MURILLO LECHUGA[1], X. LÓPEZ CAMAÑO[1], G. GUZMÁN TENORIO[1], G. ROBLES RODRÍGUEZ[1], E. ORTIZ TAPIA[1], A. PIÑA HERNÁNDEZ[1], O. ALDANA ASSAD[3], A. MEDINA RIVERA[3], A. E. RUIZ-CONTRERAS[4], M. E. RENTERÍA[5], S. ALCAUTER[1]; [1]Inst. De Neurobiología, UNAM, Queretaro, Mexico; [2]Student Undergraduate, Escuela Nacional de Estudios Superiores Juriquilla, UNAM, Queretaro, Mexico; [3]Lab. Internacional de Investigación sobre el Genoma Humano, UNAM, Queretaro, Mexico; [4]Lab. Neurogenomica Cognitiva, Fac. Psicologia, Univ. Nacional Autonoma de Mexico, Ciudad de Mexico, Mexico; [5]QIMR Berghofer Med. Res. Institute, Brisbane, QLD, Australia

Abstract: Volume, surface area, and thickness are traits of brain cortex morphology commonly studied in different populations. However, these measures are often used interchangeably, despite the cortical volume being the product of surface area and thickness, and the latter two appear to show no genetic overlap (Panizzon et al, 2009). This suggests that cortical surface and cortical thickness might be complex traits that are relevant to study independently from cortical volume, because of their different anatomical properties and different genetic influences. Still, most studies have examined the heritability and genetic correlation of these traits on populations of European and Asian ancestry, and less research on this matter has been conducted on Latin American populations. The heritability and genetic overlap of traits can differ across populations. Therefore, the present study aims to characterize the heritability and genetic relationships between global measures of the cortical surface area and cortical mean thickness in a sample of Mexican twins.

For this study, we had a sample of 95 pairs of twins (63 MZ and 32 DZ) from the Mexican twin registry (TwinsMX), who underwent MRI at the LANIREM. High-resolution T1-weighted images were processed using FreeSurfer recon-all pipeline, which produces an estimate of cortical surface area, mean thickness, and volume. The heritability and genetic cross-trait correlations of both phenotypes were estimated with a bivariate ACE model. Analyses were performed with umx, and OpenMX R v4.2.1 packages.
The heritability for the cortical surface area and mean cortical thickness was 72.93% and 22.09% respectively. On the other hand, the cross-trait genetic correlation between traits was rA = 0.45 and they had a phenotypic correlation of rP = 0.80.
The heritability of cortical surface area was similar to those found in the Vietnamese population (89%), but cortical thickness is much lower than those reported (81%). In contrast to the previous study, we found a considerable genetic and phenotypic correlation between cortical thickness and cortical surface area, showing that there is some degree of genetic overlap between the two phenotypes that explain cortical volume. Despite this overlap, it is possible that the two phenotypes have distinct genetic influences, as explained by the radial unit hypothesis of cortical development. However, these measures variate between anatomic structures. Therefore, subsequent analyses in specific cortical areas rather than global measures, and genetic variations are necessary to determine whether this genetic overlap remains, and if so, which genes could be associated with them.

Disclosures: I. Espinosa Méndez: None. **T.V. Román López:** None. **D. Ramírez González:** None. **I.C. Sanchéz Moncada:** None. **X. Díaz Téllez:** None. **C. Domínguez Frausto:** None. **V. Murillo Lechuga:** None. **X. López Camaño:** None. **G. Guzmán Tenorio:** None. **G. Robles Rodríguez:** None. **E. Ortiz Tapia:** None. **A. Piña Hernández:** None. **O. Aldana Assad:** None. **A. Medina Rivera:** None. **A.E. Ruiz-Contreras:** None. **M.E. Rentería:** None. **S. Alcauter:** None.

Poster

PSTR178. Genetic Techniques to Target and/or Manipulate Cells

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR178.29/Web Only

Topic: I.01. Molecular, Biochemical, and Genetic Techniques

Title: Neuronal signaling pathway genes as predictive biomarkers in bipolar disorder, suicidality, & treatment response: new results from Iranian samples

Authors: *S. AGHABOZORGAFJEH[1], J. KENNEDY[2], M. OMRANI[3];
[1]Ctr. for Addiction and Mental Hlth., Vaughan concord, ON, Canada; [2]Ctr. for Addiction and Mental Hlth., Toronto, ON, Canada; [3]Shahid Beheshti Univ. of Med. Sci., Tehran, Iran, Islamic Republic of

Abstract: Psychiatry in Iran has experienced an increase in bipolar disorder (BD) diagnosis and suicidality in recent years. Several pathways may lead to a bipolar diagnosis. Iranians may

experience BD differently due to sociopolitical factors and additional stressors. Wnt signaling pathway, due to its crucial role in neurodevelopment and in regulating the function and structure of the adult nervous system, has been examined in several psychiatric studies. BD often exhibits neurodevelopmental, structural, and/or functional neuronal abnormalities. We hypothesized that genetic risk factors might be more detectable given the relative homogeneity of the social stressors**.** In this study, 205 Iranian bipolar patients and 205 matched healthy controls were recruited. DNA was extracted from blood. The tetra primers-Amplification Refractory Mutation System-PCR method was used for genotyping 15 variants from 9 genes. Obtained results were confirmed by Sanger sequencing. The identified genetic variants were further investigated using bioinformatics tools such as Haploreg to assess their potential function. The findings of the study revealed associations (uncorrected) between genetic variants of *MARK2*, *BDNF*, *GSK3B*, *PPARD*, and *ADCY2* genes and certain aspects of BD. The *MARK2* rs10792421 showed an association with treatment response to lithium ($p = 0.028$), indicating its potential as a biomarker for predicting treatment outcomes. The *BDNF* Val66Met variant exhibited associations with bipolar susceptibility, suicidal behavior, and response to lithium treatment. The *GSK3B* rs334558 variant was found to be associated with patients' response to lithium treatment ($p= 0.16$), while the *PPARD* rs2267665 variant showed an association with BD susceptibility ($p=0.009$). The study also identified *ADCY2* rs2290910 as a potential biomarker for BD ($p = 0.001$), suicide tendency ($p = 0.004$), and response to lithium ($p = 0.001$), particularly in the female population. Expression of associated genes are involved in neuronal networks crucial for the neural plasticity of key brain regions, such as the hippocampus, dorsolateral prefrontal cortex, and amygdala. In-silico analysis revealed their potential regulatory role and predicted their involvement in neuronal networks, further supporting the importance of these genetic biomarkers in neuroplasticity and BD. Replication in larger patients sample in Toronto (899) is in progress. This preliminary study suggests the importance of genetic biomarkers involved in neuronal pathways in BD and related behaviours. The identification of genetic biomarkers can contribute to personalized treatment approaches and improved management of BD.

Disclosures: S. Aghabozorgafjeh: None. **J. Kennedy:** None. **M. Omrani:** None.

Poster

PSTR178. Genetic Techniques to Target and/or Manipulate Cells

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR178.30/VV32

Topic: I.01. Molecular, Biochemical, and Genetic Techniques

Title: Cross disorder patient and mechanistic stratification using combinatorial analyses

Authors: *S. HOGG, **K. CHOCIAN**, S. DAS, A. MALINOWSKI, K. TAYLOR, S. BEAULAH;
PrecisionLife, Long Hanborough, United Kingdom

Abstract: Central nervous system (CNS) disorders encompass a broad spectrum of conditions that share some common characteristics including clinical phenotypes such as cognitive dysfunction, sleep disturbance and mood changes. This suggests the existence of shared pathophysiological mechanisms that can provide insights into development of common therapies. Genetic studies aiming to identify common mechanisms across CNS disorders are constrained by the limitations of GWAS approaches which typically identify isolated genetic variants with high prevalence. The key to understanding complex, multi-symptomatic CNS syndromes influenced by multiple genetic loci, epidemiological and/or environmental factors is to find combinations of these factors in sub-cohorts with distinct characteristics. We use a hypothesis-free method for the detection of combinations of genetic factors that together are strongly associated with variations in disease risk, symptoms, prognoses, and other disease-associate variables in patient subgroups. Patient stratification insights generated by these combinatorial disease signatures enable identification of mechanistic subgroups that are common to multiple indications. The PrecisionLife platform was used to compare disease to control and identify common genetic variants across a wide range of CNS indications; subsequent enrichment analyses were performed to determine pathways and biological processes that are significantly associated with the genetic variants identified for each indication. This enabled us to identify biological mechanisms that are impacted across CNS diseases. Our results identified multiple common biological processes that are significantly affected in several CNS diseases such as neurogenesis, neurotransmission, cell death signalling and lipoprotein metabolism; 78 disease-associated genes were identified in two or more different CNS indications, including 22 enzymes, 10 transcription factors and 9 transporters. Combinatorial analyses can stratify heterogenous patient populations with complex pathologies to identify mechanistically driven stratification insights that classify patients beyond traditional syndromal diagnostic categories. Our analyses indicate that subgroups of patients with CNS diseases have common pathophysiological drivers that may contribute to specific common clinical manifestations; the patients in these subgroups can be identified using combinatorial genetic patient stratification biomarkers, to enable a targeted precision approach to symptoms occurring across multiple CNS disorders.

Disclosures: **S. Hogg:** A. Employment/Salary (full or part-time); PrecisionLife. **K. Chocian:** A. Employment/Salary (full or part-time); PrecisionLife. **S. Das:** A. Employment/Salary (full or part-time); PrecisionLife. **A. Malinowski:** A. Employment/Salary (full or part-time); PrecisionLife. **K. Taylor:** A. Employment/Salary (full or part-time); PrecisionLife. **S. Beaulah:** A. Employment/Salary (full or part-time); PrecisionLife.

Poster

PSTR179. Systems Biology and Multiomics Approaches

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR179.01/VV33

Topic: I.02. Systems Biology and Bioinformatics

Support: JST ERATO grant number JPMJER2001
the Science and Technology Platform Program for Advanced Biological Medicine (AMED/MEXT to H.R.U.)
JSPS KAKENHI grant-in-aid for scientific research (S) (H.R.U., grant number JP18H05270)
JSPS KAKENHI grant-in-aid for scientific research (C) (to K.M., grant number 20K06885)
a Grant-in-Aid from the Human Frontier Science Program (to H.R.U.)
JST [Moonshot R&D] (to K.M., grant number JPMJMS2023)
MEXT Quantum Leap Flagship Program (MEXT QLEAP) (to H.R.U., grant number JPMXS0120330644)
JSPS KAKENHI grant-in-aid for Early-Career Scientists (grant number 20K16498)

Title: Whole-brain Cellome-Wide Association Study (CWAS) reveals neurodegenerative trajectories in a 3D cellular landscape

Authors: *T. T. MITANI[1,2], K. YAMAURA[1,2], K. MATSUMOTO[1,3], E. A. SUSAKI[5,1], H. R. UEDA[1,4,2];
[1]RIKEN Ctr. for Biosystems Dynamics Res., Suita-shi, Japan; [2]Osaka university, Suita-shi, Japan; [3]The Univ. of Tokyo, Tokyo, Japan; [4]The Univ. of Tokyo, Suita-shi, Japan; [5]Juntendo university, Tokyo, Japan

Abstract: As societies age, the rise of neurodegenerative diseases like Alzheimer's requires preemptive medicine for early detection and preventive treatment. The key to discovering such effective molecular targets is to implement a comprehensive all-cell screening scheme to identify disease-affected cellular positions and the surrounding microenvironment, even at the earliest stages of these diseases. Here, we present a novel approach, CWAS (whole body/organ cellome-wide association studies), which integrates and analyzes spatio-temporal cellular information across the entire mouse brain using tissue-clearing imaging technologies. This approach reveals previously unreported cases and the non-linear effects of aging on neuronal loss during disease progression at different stages of life. In addition, our time-series data-driven analysis in an Alzheimer's mouse model challenges the traditional amyloid hypothesis by suggesting that amyloid deposition and neurodegeneration may occur simultaneously. We also reconstruct the 3D trajectory of pathological progression and successfully build a 3D disease atlas depicting the spatio-temporal expansion of neurodegenerative niches, including highly aggregated microglia and neuronal loss. This strategy allows us to visualize cellular-level changes in neurodegenerative diseases in three-dimensional space and in a sequence that reflects disease progression, as a 'pseudo-disease trajectory'. Our findings not only establish the transomics concept of cellomics, but also signal a potential paradigm shift in neuropathology, guiding future research and treatment strategies.

Disclosures: T.T. Mitani: None. **K. Yamaura:** None. **K. Matsumoto:** None. **E.A. Susaki:** None. **H.R. Ueda:** None.

Poster

PSTR179. Systems Biology and Multiomics Approaches

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR179.02/VV34

Topic: I.02. Systems Biology and Bioinformatics

Support: JST ERATO grant number JPMJER2001
the Science and Technology Platform Program for Advanced Biological Medicine (AMED/MEXT to H.R.U.)
JSPS KAKENHI grant-in-aid for scientific research (S) (H.R.U., grant number JP18H05270)
JSPS KAKENHI grant-in-aid for scientific research (C) (to K.M., grant number 20K06885)
a Grant-in-Aid from the Human Frontier Science Program (to H.R.U.)
JST [Moonshot R&D] (to K.M., grant number JPMJMS2023)
MEXT Quantum Leap Flagship Program (MEXT QLEAP) (to H.R.U., grant number JPMXS0120330644)

Title: Wise hypothesis for synaptic function during the sleep-wake cycle

Authors: *F. KINOSHITA[1], R. G. YAMADA[1], K. MATSUMOTO[2], H. R. UEDA[3,2];
[1]Osaka Univ., Suita, Japan; [2]the Univ. of Tokyo, Tokyo, Japan; [3]Biosystem dynamics Res. Ctr., Riken, Suita, Japan

Abstract: Much is still unknown about the relationship between learning rules and the synaptic-weight dynamics in cortical neuron networks during the sleep and wake cycle. The synaptic homeostasis hypothesis proposes that synaptic weight increases in the wake and decreases during sleep, which can underlie the slow dynamics of the sleep and wake cycle (G. Tononi et al., 2005). However, such synaptic-weight dynamics contradict the fast electrophysiological activity observed during sleep because the fast activity increases synaptic weight and induces the sleep-like synchronized activity of cortical neurons. We studied the role of synaptic plasticity in the sleep-wake cycle by incorporating learning rules into computational models in the following steps. First, we assumed learning rules and investigated synaptic-weight dynamics by assessing the effects of sleep-like and wake-like spike trains on the Ca2+-based plasticity model. Second, we constructed network models of autonomously firing neurons with randomly assigned parameters and simulated gradually changing the conductance of specific ion channels and synaptic receptors. We then selected models that exhibited the transition from wake-like to sleep-like states or vise-versa. In addition, we incorporated a synaptic learning rule and a model of phosphorylation hypothesis into the network to investigate the role of synaptic plasticity in the sleep-wake cycle regulated by multistep auto-phosphorylation of a kinase. We found that the synaptic weight decreased in the wake-like state while it increased in the sleep-like state under Hebbian and classical spike-timing-dependent plasticity, especially at low frequencies. We call such synaptic-weight dynamics the WISE (Wake Inhibition and Sleep Excitation) hypothesis. The WISE hypothesis was robust to changes in the network connection, spike pattern, and the sleep-wake cycle.

Disclosures: **F. Kinoshita:** None. **R.G. Yamada:** None. **K. Matsumoto:** None. **H.R. Ueda:** None.

Poster

PSTR179. Systems Biology and Multiomics Approaches

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR179.03/VV35

Topic: I.02. Systems Biology and Bioinformatics

Support: JST ERATO grant number JPMJER2001
the Science and Technology Platform Program for Advanced Biological Medicine (AMED/MEXT to H.R.U.)
JSPS KAKENHI grant-in-aid for scientific research (S) (H.R.U., grant number JP18H05270)
JSPS KAKENHI grant-in-aid for scientific research (C) (to K.M., grant number 20K06885)
a Grant-in-Aid from the Human Frontier Science Program (to H.R.U.)
JST [Moonshot R&D] (to K.M., grant number JPMJMS2023)
MEXT Quantum Leap Flagship Program (MEXT QLEAP) (to H.R.U., grant number JPMXS0120330644)

Title: Whole brain activity atlas of circadian rhythm: A quantitative analysis with tissue clearing, 3D immunostaining, and 3D imaging

Authors: *K. YAMASHITA[1], K. MATSUMOTO[1,2], S. Y. YOSHIDA[1,2], H. R. UEDA[1,2];
[1]RIKEN Lab. for Synthetic Biol., Suita-shi, Osaka, Japan; [2]Univ. of Tokyo, Tokyo, Japan

Abstract: When evaluating the response to certain medications in specific brain regions, experiments are often performed at different time points throughout the day. However, it can be difficult to distinguish whether the observed response is due to the effect of the medication or circadian changes. Therefore, it is crucial to establish the baseline neuronal activities in the whole brain over a 24-hour period to understand the circadian changes comprehensively. The brain consists of various interconnected regions which form complex networks. Recent advances in tissue clearing and three-dimensional (3D) imaging techniques offer the potential to study the organized brain functions as a whole, specifically targeting immediate early gene (IEG) expression throughout the entire brain. Unlike IEGs that are localized in specific regions, such as the Arc gene expressed in cortex and hippocampus, c-Fos is known to be widely expressed throughout the brain. We have developed a technique for whole-brain c-Fos immunostaining, imaging, and analysis using CUBIC (Clear, Unobstructed Brain Imaging Cocktails and Computational Analysis), which enables easy and unbiased analysis of whole-brain neuronal activities at single-cell resolution in an easy and non-biased manner (Reference: Susaki, et al. Cell 2014). While the circadian rhythm of c-Fos in the suprachiasmatic nucleus of the hypothalamus (SCN), the center of the circadian clock in the body, has been commonly reported,

reports on circadian changes in other regions are limited. Here, we focused on investigating the circadian changes of c-Fos expression across all brain regions. Our methods involved placing mice in a dark-dark (DD) condition following exposure to a regular light-dark (LD) condition and performing time-series sampling of their brains. We then captured whole-brain c-Fos immunostaining images at single-cell resolution using CUBIC with a light-sheet microscopy and quantified them in each region using a brain atlas (Ref: Murakami, et al. Nature Neuroscience 2018). Our findings revealed the circadian changes of c-Fos expression not only in the SCN but also in other regions. This comprehensive understanding of circadian dynamics of neural activities across diverse brain regions has the potential to contribute to the development of more precise brain researches.

Disclosures: K. Yamashita: None. **K. Matsumoto:** A. Employment/Salary (full or part-time); CUBICStars. **S.Y. Yoshida:** A. Employment/Salary (full or part-time); CUBICStars. **H.R. Ueda:** None.

Poster

PSTR179. Systems Biology and Multiomics Approaches

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR179.04/VV36

Topic: I.02. Systems Biology and Bioinformatics

Support: JST ERATO grant number JPMJER2001
the Science and Technology Platform Program for Advanced Biological Medicine (AMED/MEXT to H.R.U.)
JSPS KAKENHI grant-in-aid for scientific research (S) (H.R.U., grant number JP18H05270)
JSPS KAKENHI grant-in-aid for scientific research (C) (to K.M., grant number 20K06885)
a Grant-in-Aid from the Human Frontier Science Program (to H.R.U.)
JST [Moonshot R&D] (to K.M., grant number JPMJMS2023)
MEXT Quantum Leap Flagship Program (MEXT QLEAP) (to H.R.U., grant number JPMXS0120330644)

Title: Applications of the whole organ/body atlas of mice with a single-cell resolution

Authors: *S. Y. YOSHIDA[1,2], T. T. MITANI[1,3], K. MATSUMOTO[2,1], H. R. UEDA[2,1]; [1]RIKEN Ctr. For Biosystems Dynamics Res., Suita-shi/ osaka, Japan; [2]Grad. Sch. of Med., The Univ. of Tokyo, Tokyo, Japan; [3]Grad. Sch. of Med., Osaka university, Suita-shi/ osaka, Japan

Abstract: A range of issues, including cancer and drugs, may damage systemic organs such as the lungs, kidneys, heart, and liver. By sectioning the area of interest and only partially observing it, conventional pathological examinations of organs have been performed. Observing the entire organ at single-cell resolution will make it possible to count the number of cancer cells that have

spread and are still present within an organ, as well as to assess drug-induced organ damage with great sensitivity and accuracy. Tissue clearing and light-sheet microscopy, as represented by the CUBIC (clear, unobstructed Brain Imaging Cocktails and Computational Analysis) approach, have recently made whole organ observation feasible (Reference: Susaki et al. Cell 2004). This method enabled the creation of a mouse brain atlas with single-cell resolution (Ref. Murakari et al. Nature Neuroscience. 2018, Matsumoto et al. Nature Protocols 2019). This brain atlas made it possible to analyze changes in all areas of the brain (Ref. Mano et al Cell Reports Methods 2021), however, there were no single-cell resolution atlases available for any organs other than the brain. To provide the foundation for future organ analysis, single-cell resolution atlases of numerous organs were first made. We prepared organs of 8-week-old mice using the CUBIC method. The lung, kidney, heart, liver, bladder, testis, thyroid, salivary glands, and pancreas were cleared, and took images with a custom-made light sheet microscope. We counted the cells automatically using GPU. Now we show the examples of application of whole cell analysis; we observed kidney development, kidney damage caused by cisplatin, and lung damage caused by LPS. We segmented the organs and analyzed the perturbations in each region. We also attempted to define new regions of organs by analyzing region-independent perturbations. In addition, we created a whole-body atlas of newborn mice with single-cell resolution. Whole body clearing using the customized CUBIC revealed more than 400 million cells. These cells were also analyzed by region. Additionally, immunostaining showed the quantity and distribution of macrophages throughout the body. Due to their ability to analyze at the cellular level, these atlases may be crucial in the study of organs and the entire body.

Disclosures: **S.Y. Yoshida:** A. Employment/Salary (full or part-time); CUBICStars. **T.T. Mitani:** None. **K. Matsumoto:** A. Employment/Salary (full or part-time); CUBICStars. **H.R. Ueda:** None.

Poster

PSTR179. Systems Biology and Multiomics Approaches

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR179.05/VV37

Topic: I.02. Systems Biology and Bioinformatics

Support: JST [Moonshot R&D] (to K.M., grant number JPMJMS2023)
JST ERATO grant number JPMJER2001
the Science and Technology Platform Program for Advanced Biological Medicine (AMED/MEXT to H.R.U.)
JSPS KAKENHI grant-in-aid for scientific research (S) (H.R.U., grant number JP18H05270)
a Grant-in-Aid from the Human Frontier Science Program (to H.R.U.)
MEXT Quantum Leap Flagship Program (MEXT QLEAP) (to H.R.U., grant number JPMXS0120330644)
JSPS KAKENHI grant-in-aid for scientific research (C) (to K.M., grant number 20K06885)

Title: Development of the simple tissue clearing protocol

Authors: *K. MATSUMOTO[1,2,3], F. AKIYAMA[2,4], H. R. UEDA[3,2];
[1]Tokyo Univ., Tokyo, Japan; [2]Ctr. for Biosystems Dynamics Res., RIKEN, Suita, Japan; [3]Dept. of Systems Pharmacol., The Univ. of Tokyo, Tokyo, Japan; [4]Nagasaki Univ., Nagasaki, Japan

Abstract: Recent developments of tissue clearing technology and 3D imaging and 3D image analysis technology using light sheet microscopy have made it possible to easily observe all cells, structures, and drug localization in tissues or organs. When this technology is applied to biological research or drug discovery research that requires a large number of samples, at least several tens of samples must be handled at the same time. However, many protocols process a single sample in a conical tube, and even simple processes such as reagent exchange and washing processes require enormous amounts of labor. Therefore, we developed a high-throughput, simple, and space-saving tissue clearing protocol using multi-well plates. Among the various transparency reagents and methods, we chose to use CUBIC (Clear, Unobstructed Brain / Body Imaging Cocktails and Computational analysis), which is a passive tissue clearing method, water soluble which compatible with the plastic of multi-well plates, and easy to handle. An aspirator was used to simplify solution exchange, an inner device was designed to fit into a 6-well plate with a covered area for aspiration to prevent tissue damage when aspirating solutions and a light path for photographing samples with a light sheet microscope. In addition, in order to easily take images with a light-sheet microscope for cleared samples, we also optimized the method of agarose gel embedding with this device. We confirmed and optimized the each conditions using this device performances of the delipidation efficiency, solution exchange efficiency, stainability of nuclear staining, and transparency of the whole brain and other organs, and we developed a parallel and simple tissue clearing protocol for mouse organs.

Disclosures: **K. Matsumoto:** A. Employment/Salary (full or part-time); CUBICStars.co.,. **F. Akiyama:** None. **H.R. Ueda:** A. Employment/Salary (full or part-time); CUBICStars.co.,.

Poster

PSTR179. Systems Biology and Multiomics Approaches

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR179.06/VV38

Topic: I.02. Systems Biology and Bioinformatics

Support: AMED JP20gm6210027, JP21wm0425003
KAKENHI 22H02824, 22H04926
UTEC-UTokyo
The Takeda Science Foundation
Nakatani foundation
Mochida Memorial Foundation

Title: Cubic-histovision2.0: expanded volumetric staining through phase separation and compound optimization

Authors: *E. A. SUSAKI[1,2], Y. SAITO[1], S. YOSHIDA[2], C. SHIMIZU[2], M. KURODA[3], H. R. UEDA[2,4];
[1]Dept. of Biochem. and Systems Biomedicine, Grad. Sch. of Med., Juntendo Univ., Tokyo, Japan; [2]Lab. for Synthetic Biol., RIKEN Ctr. for Biosystems Dynamics Res., Osaka, Japan; [3]Intl. Res. Ctr. for Neurointelligence (WPI-IRCN), [4]Dept. of Systems Pharmacology, Grad. Sch. of Med., The Univ. of Tokyo, Tokyo, Japan

Abstract: The development of tissue clearing techniques has led to an expansion of their applications. Histological labeling has several advantages over genetic labeling, while inadequate penetration of staining probes is a drawback. To address this issue, we have provided CUBIC-HistoVIsion (Susaki et al. Nature Communications 2020, 11:1982) as a rational design for effective 3D staining protocols based on the physicochemical properties of biological tissue as an electrolyte gel. However, this protocol involves multiple stages and is time-consuming, typically requiring 2-4 weeks for staining alone.
Here, we describe the development of an advanced CUBIC-HistoVIsion2.0 protocol intended to enhance 3D staining in a short time and in large tissue samples. Assuming the reaction-diffusion process of 3D staining, we focused on two crucial parameters: the initial concentration of staining probes and the modulation of probe-target binding efficiency. We developed HV-LLPS (liquid-liquid phase separation), a technology that condenses staining probes 5-10 times higher (50-100 μg/mL) than the current protocol (typically 10 μg/mL). This is achieved by surrounding the staining buffer and the sample (the aqueous phase) with a reagent that does not mix with water (the organic phase). By utilizing HV-LLPS, we were able to obtain a very high concentration of the probes while using a minimal amount of staining buffer per sample. Furthermore, by screening over 500 compounds, we identified a set of HV-additives that enhance probe penetration in 3D tissue samples through modulation of probe-tissue binding. The resultant compound enhanced probe penetration significantly more than the chemicals used in the initial protocol. The newly developed protocol provides a considerably shorter (typically one week for staining) and scalable (enabling stable staining of the entire rat brain) one-step protocol. We presented cases of multiple whole mouse brain stainings, as well as whole P0 mouse body staining (targets to be stained inside the cranium) and whole rat brain staining.
Overall, the implementation of our advanced CUBIC-HistoVIsion2.0 protocol provides practical solutions to surmount the burden of 3D histological staining and imaging, which will be an indispensable instrument for future comprehensive tissue and organ analysis. In addition, our research contributes to demonstrating the proof-of-concept of the theoretically assumed essential 3D staining parameters and their experimental implementation using simple and replicable technologies.

Disclosures: **E.A. Susaki:** A. Employment/Salary (full or part-time); CUBICStars, Inc.. C. Other Research Support (receipt of drugs, supplies, equipment or other in-kind support); Tokyo Chemical Industry. **Y. Saito:** None. **S. Yoshida:** A. Employment/Salary (full or part-time); CUBICStars, Inc.. **C. Shimizu:** None. **M. Kuroda:** None. **H.R. Ueda:** A. Employment/Salary (full or part-time); CUBICStars, Inc.. C. Other Research Support (receipt of drugs, supplies, equipment or other in-kind support); Tokyo Chemical Industry. E. Ownership Interest (stock,

stock options, royalty, receipt of intellectual property rights/patent holder, excluding diversified mutual funds); CUBICStars, Inc.

Poster

PSTR179. Systems Biology and Multiomics Approaches

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR179.07/VV39

Topic: I.02. Systems Biology and Bioinformatics

Title: Highly optimized whole-oragan immunostaining and nuclear staining methods and its applications

Authors: *Y. OTSUKI[1], K. MATSUMOTO[2,1,3], Y. OKABE[1], S. Y. YOSHIDA[3,1,2], E. A. SUSAKI[4,1], H. R. UEDA[2,1,3];
[1]CUBICStars, Tokyo, Japan; [2]Univ. of Tokyo, Tokyo, Japan; [3]RIKEN Ctr. For Biosystems Dynamics Res., Suita-shi/ osaka, Japan; [4]Juntendo Univ., Tokyo, Japan

Abstract: Examining entire mammalian organs as functional assemblies of individual cells is an important goal of biology and medicine (Reference: Tainaka et al. Cell Repots 2018). Because brains have complex networks of neurons called connectome, it is important to observe the neural connections throughout the organ in three dimensions. Furthermore, neurons, including excitatory and inhibitory neurons, have various characteristics, and distinguishing between them leads to an understanding of the complex functions of the brain. In recent years, combining tissue clearing methods and three-dimensional staining methods with light-sheet microscopy has made it possible to perform three-dimensional imaging of entire organs such as brains. The representative method is CUBIC (clear, unobstructed Brain Imaging Cocktails and Computational Analysis), which is widely used because it is compatible with the preservation of fluorescent proteins, safety, and immunostaining (Reference: Susaki et al. Cell 2004, Susaki et al. Nature communications 2020). Because of the many precautions required in the procedure for three-dimensional immunostaining of whole organs, the introduction of this method at other research facilities sometimes failed. In addition, the optimum conditions varied depending on the antibodies, therefore it is difficult to get sufficient staining results. We have developed a three-dimensional immunostaining kit (CUBIC-HV™2 3D tissue staining kit) that can stain stably in any laboratory. We also developed protocols using this kit that achieves uniformity of staining, speed of antibody penetration, and high signal-to-noise ratio by examining the appropriate additives and temperature of staining for each antibody. Using this kit (CUBIC-HV™2 3D tissue staining kit) and the protocols, it will be easy for other research facilities to introduce three-dimensional immunostaining of whole organs, which is expected to further develop a wide range of research, including in the field of neuroscience.

Disclosures: **Y. Otsuki:** A. Employment/Salary (full or part-time); CUBICStars. **K. Matsumoto:** A. Employment/Salary (full or part-time); CUBICStars. **Y. Okabe:** A. Employment/Salary (full or part-time); CUBICStars. **S.Y. Yoshida:** A. Employment/Salary (full

or part-time); CUBICStars. **E.A. Susaki:** A. Employment/Salary (full or part-time); CUBICStars. **H.R. Ueda:** A. Employment/Salary (full or part-time); CUBICStars.

Poster

PSTR179. Systems Biology and Multiomics Approaches

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR179.08/VV40

Topic: I.02. Systems Biology and Bioinformatics

Support: AMED JP20gm6210027
AMED JP22ama221517
AMED JP21ak0101181
AMED JP21wm0425003
CREST JPMJCR20E4
JSPS KAKENHI 22H02756
JSPS KAKENHI 22H02824
JSPS KAKENHI Advanced Bioimaging Support JP22H04926
Operating Costs Subsidies for Private Universities
UTEC-UTokyo
Takeda Science Foundation
Nakatani foundation for advancement of measuring technologies in biomedical engineering
Mochida Memorial Foundation for Medical and Pharmaceutical Research

Title: Descspim: affordable and easy-to-build light-sheet microscopy for tissue clearing technique users

Authors: *T. OMURA[1,2,3,4], K. OTOMO[1,2,5,6], Y. NOZAWA[1], Y. SAITO[2], Y. WATAKABE[5,6], M. SUZUKI[7], S. YAGISHITA[8], A. HAMADA[7,8], H. UCHIDA[9], K. TAINAKA[9,10], E. A. SUSAKI[1,2,10];
[1]Biochem. II, Juntendo Univ., Tokyo, Japan; [2]Dept. of Biochem. and Systems Biomedicine, Juntendo Univ. Grad. Sch. of Med., Tokyo, Japan; [3]Dept. of Neurosurg., Univ. of Tokyo, Tokyo, Japan; [4]Dept. of Neurosurg. and Neuro-Oncology, Natl. Cancer Ctr. Hosp., Tokyo, Japan; [5]Div. of Biophotonics, Natl. Inst. for Physiological Sci., [6]Biophotonics Res. Group, Exploratory Res. Ctr. on Life and Living Systems, Natl. Inst. of Natural Sci., Aichi, Japan; [7]Dept. of Pharmacol. and Therapeutics, Fundamental Innovative Oncology Core, [8]Div. of Mol. Pharmacol., Natl. Cancer Ctr. Res. Inst., Tokyo, Japan; [9]Dept. of Syst. Pathology for Neurolog. Disorders, Brain Res. Inst., Niigata Univ., Niigata, Japan; [10]Lab. for Synthetic Biol., RIKEN Ctr. for Biosystems Dynamics Res., Osaka, Japan

Abstract: Introduction: In the field of biomedical research, light-sheet fluorescence microscopy (LSFM) has revolutionized how we visualize and comprehend biological specimens. However, its widespread adoption has been hindered due to high costs and the technical expertise required.

To address this, we developed a novel, cost-effective, and user-friendly solution: the desktop-equipped SPIM for cleared specimens (descSPIM)[1]. This do-it-yourself light-sheet microscopy system democratizes access to LSFM, enabling routine three-dimensional imaging of cleared samples within minutes. Results: The descSPIM system employs a one-sided light-sheet illumination with minimum configuration. Lights-sheet formed by a collimated lens and a cylindrical lens, and the detection path comprises a 2X objective lens, a tube lens, and a CMOS camera, with an effective integration magnification of 1X. A z-stack image was rapidly collected as time-lapse (xy-t) data by the continuous moving of the actuators, and then the data was converted into xy-z format.For practical use, we adopted two types of cylindrical lenses to create wider or axially finer illumination. Full-FOV (FF) illumination mode enables an axial spatial resolution of 26 ± 0.5 μm. The other cylindrical lens with a shorter focal length yielded fine-axial (FA) illumination with the axial spatial resolution of 7.2 ± 0.3 μm. We've demonstrated descSPIM's efficacy by imaging a CUBIC-cleared mouse hemisphere, whole brain, a 2 mm-thick mouse brain slice, and tissues derived from human cancer patients. Furthermore, we've innovated a method to compensate for signal intensity differences in unidirectionally illuminated light-sheet microscopy, using Advanced Normalization Tools (ANTs) for successful fusion of double-sided images at sub-cellular resolution. Discussion: descSPIM's basic configuration is simple and highly customizable, making it an ideal introductory light-sheet fluorescence microscope for researchers transitioning from 2D to 3D imaging.
[1] Otomo, K., Omura, T., Nozawa, Y., Saito, Y., & Susaki, E. A. (2023). descSPIM: Affordable and Easy-to-Build Light-Sheet Microscopy for Tissue Clearing Technique Users. https://doi.org/10.1101/2023.05.02.539136

Disclosures: **T. Omura:** None. **K. Otomo:** None. **Y. Nozawa:** None. **Y. Saito:** None. **Y. Watakabe:** None. **M. Suzuki:** None. **S. Yagishita:** None. **A. Hamada:** None. **H. Uchida:** None. **K. Tainaka:** None. **E.A. Susaki:** A. Employment/Salary (full or part-time); CUBICStars Inc.

Poster

PSTR179. Systems Biology and Multiomics Approaches

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR179.09/VV41

Topic: I.02. Systems Biology and Bioinformatics

Title: Clearing techniques for a high-throughput whole mouse brain imaging platform

Authors: *E. PETERSON[1], M. TAORMINA[1], D. TOGLIA[2], J. ROHDE[2], A. GLASER[2], J. CHANDRASHEKAR[2], C. M. PAGAN[1], J. WATERS[1];
[1]Allen Inst. for Brain Sci., Seattle, WA; [2]Allen Inst. for Neural Dynamics, Seattle, WA

Abstract: Whole mouse brain imaging enables the ability to undertake large-scale studies, such as the screening of viral genetic tools, connectivity of brain regions, characterization of transgenic mouse lines, and the development of anatomical atlases. Previous serial-sectioning

two-photon tomography studies generated data that led to the development of widely used resources in the neuroscience community. With advances in both tissue clearing and imaging, adopting a lightsheet fluorescence imaging strategy promises improvements in imaging time, sampling density, and spectral multiplexing. However, whole brain imaging requires opaque brain tissue to be optically cleared prior to imaging. With a multitude of clearing methods available, each yielding different results, careful evaluation is necessary to select the most suitable method for a large-scale imaging platform. There are two categories for clearing tissue, utilizing either organic solvents or aqueous detergents. Solvent- based methods require dehydration during delipidation, causing tissue contraction and loss of protein fluorescence. Aqueous clearing methods maintain original tissue size and preserve native fluorescence, making them good candidates for these types of experimental modalities. We tested four different aqueous based clearing methods across twenty-four whole mouse brains to determine which method is the most suitable for our large-scale platform. Three of the four methods tested were aqueous based passive clearing, and the other was an aqueous based electrophoretic clearing method. Twelve of these brains had endogenous cytosolic labeling and were from the same breeding group (Rbp4-Cre_KL100/wt;Ai193(TICL-EGFP-ICF-tdT)-hyg/wt), the other twelve had endogenous nuclear labeling. All twenty-four mouse brains were imaged on the SmartSPIM Imaging Platform from LifeCanvas with standardized acquisition parameters. The different clearing methods were compared based on image sharpness and their ability to preserve signal and reduce background across the brain. One method, LifeCanvas Passive Clearing, yielded an expanded sample causing low signal preservation. Next, we tested EZ Clear, which performed poorly in signal preservation and image sharpness. We also tested an unpublished method, which had good signal preservation but poor image sharpness due to inconsistencies in the refractive index. We found that aqueous electrophoretic clearing produced superior data quality, making it the most suitable for large-scale studies such as screening viral genetic tools, characterizing transgenic mouse lines, and developing anatomical atlases.

Disclosures: E. Peterson: None. **M. Taormina:** None. **D. Toglia:** None. **J. Rohde:** None. **A. Glaser:** None. **J. Chandrashekar:** None. **C.M. Pagan:** None. **J. Waters:** None.

Poster

PSTR179. Systems Biology and Multiomics Approaches

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR179.10/VV42

Topic: I.02. Systems Biology and Bioinformatics

Title: Bridging Function and Transcriptomics: Experimental methods to link gene expression to in vivo physiology in mice

Authors: *C. L. BONATTO PAESE, J. KIM, E. PETERSON, H. SCHRYVER, C. BERRY, S. MCCULLOCH, O. ZOBEIRI, M. DAVIS, L. SUAREZ, J. WILKES, M. TAORMINA, S. CALDEJON, A. WILLIFORD, J. MILLER, J. WATERS, S. OLSEN, M. GARRETT, A.

ARKHIPOV, P. GROBLEWSKI;
Allen Inst., Seattle, WA

Abstract: The brain exhibits an exceptional level of complexity and diversity, requiring precise regulation of gene expression to maintain its intricate network of neural circuits. Gene expression patterns in the brain are highly dynamic and can vary across different regions, cell types, and developmental stages. To understand more about these dynamic patterns, we have developed a platform to link functional imaging data collected in vivo with spatial transcriptomics data collected ex vivo. Here we describe the development and validation of experimental methodology to perform these complex experiments in a reliable, reproducible, and efficient manner. We use longitudinal in vivo 2-photon calcium imaging in transgenic mice to measure the activity of neurons in visual cortex during visual perception and behavior. To map more highly resolved transcriptomic cell types, the brain of the mice are sectioned, cleared, expanded, and undergo hybridization chain reaction (HCR). The use of thick tissue sections (350um before expansion) poses many challenges for a standard HCR protocol. Further, probing for enough gene transcripts to map on to existing cell type taxonomies requires performing multiple rounds of HCR, which can take weeks with standard protocols. Improvements in our HCR protocol have been achieved through several strategies. We have piloted a novel reversible HCR technique, which to date has not been tested in thick tissue mouse samples. With this approach, up to 20 genes are expected to be detected and imaged in five days. We have improved fixation techniques, decreased background signal, thereby increasing our gene expression signal significantly. Co-registration of expanded lightsheet imaging datasets back onto the previously collected in vivo 2-photon calcium imaging datasets is a challenging aspect of our experimental workflow, made easier by the presence of robustly identifiable fiducial markers. We are currently exploring multiple methods for labeling vasculature, for example with the systemic injection of fluorescently labeled compounds, as well as other means of marking the regions where functional imaging occurred in vivo, such as near-infrared branding. The combination of optimized HCR and vasculature labeling will help us to link the brain structure with its function, and by identifying cell types and their molecular patterns we will be able to explore the molecular landscape of the brain with unprecedented precision. Finally, these datasets will help us to elucidate gene expression patterns, neuronal cell types, neuronal connectivity, and molecular dynamics underlying neural development, plasticity, and disease pathology.

Disclosures: **C.L. Bonatto Paese:** A. Employment/Salary (full or part-time); Allen Institute. **J. Kim:** None. **E. Peterson:** None. **H. Schryver:** None. **C. Berry:** None. **S. McCulloch:** None. **O. Zobeiri:** None. **M. Davis:** None. **L. Suarez:** None. **J. Wilkes:** None. **M. Taormina:** None. **S. Caldejon:** None. **A. Williford:** None. **J. Miller:** None. **J. Waters:** None. **S. Olsen:** None. **M. Garrett:** None. **A. Arkhipov:** None. **P. Groblewski:** None.

Poster

PSTR179. Systems Biology and Multiomics Approaches

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR179.11/VV43

Topic: I.02. Systems Biology and Bioinformatics

Title: Bridging Function and Transcriptomics: A high-throughput, flexible quality control system for multiple data modalities in an open-science environment

Authors: *S. MCCULLOCH[1], C. ROLL[1], S. M. SEID[1], S. CALDEJON[1], N. ORLOVA[1], A. LEON[1], M. S. ALOI[1], C. BERRY[1], M. DAVIS[1], J. KIM[1], S. R. OLSEN[1], P. A. GROBLEWSKI[2], M. GARRETT[3];
[1]Allen Inst., Seattle, WA; [2]Allen Inst. For Brain Sci., Seattle, WA; [3]Allen Inst. for Brain Sci., Seattle, WA

Abstract: Data quality is vital in open-science research. Incorporating robust and timely quality control (QC) methods into the research environment facilitates rapid iteration, upholds rigorous standards, and provides the tools necessary for optimizing data preservation and provenance. We describe a platform that applies a robust QC framework across diverse data modalities and experimental procedures as part of the Allen Brain Observatory. Our QC system has been used in the generation of multiple open-access datasets, including the Visual Coding 2P and Visual Behavior 2P projects (brain-map.org/explore/circuits). Here we describe the extension and modernization of our QC infrastructure to support new data types and experiments combining longitudinal in vivo 2-photon calcium imaging during behavior with post-hoc spatial transcriptomics with multiplexed fluorescence in-situ hybridization (mFISH) and lightsheet imaging. These multi-modal, complex experiments involve numerous data streams and extensive metadata about experimental parameters and data organization. A few of the varied data streams include imaging time-series and volumes, task specific behavior like lick and running data, and acquisition metadata such as data synchronization and frame timing. Each data stream is independently evaluated with customized metrics and visuals to assess issues specific to that data type. Rapid turnaround in generation and review of QC reports is particularly important in the adaptive experimental paradigms central to combining longitudinal in vivo 2-photon calcium imaging, mFISH and lightsheet imaging. By incorporating next-day QC review of acquired data, we provide critical information necessary for experimental decisions. These include tracking task performance to inform optimal training progression, verifying field-of-view matches to enable cell tracking over weeks of imaging, and analyzing imaging quality and hardware performance to ensure consistent, high quality data acquisition. To best support these diverse projects and multi-modal research efforts, the QC system needs to be easily extendable, customizable, and able to provide granular annotations. By incorporating these practices, the platform fosters trust, instills confidence, and elevates overall data quality in neuroscience research. By outlining the diverse metrics we employ, and our strategy for QC organization, we aim to stimulate ideas and offer a blueprint for researchers to enhance their own QC processes. Future work includes efforts to transition to a cloud-based architecture to provide public insight into data quality efforts.

Disclosures: S. McCulloch: None. **C. Roll:** None. **S.M. Seid:** None. **S. Caldejon:** None. **N. Orlova:** None. **A. Leon:** None. **M.S. Aloi:** None. **C. Berry:** None. **M. Davis:** None. **J. Kim:** None. **S.R. Olsen:** None. **P.A. Groblewski:** None. **M. Garrett:** None.

Poster

PSTR179. Systems Biology and Multiomics Approaches

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR179.12/VV44

Topic: I.02. Systems Biology and Bioinformatics

Title: Bridging Function and Transcriptomics: An open-source cloud-based data processing pipeline for 2-photon and lightsheet imaging data

Authors: *M. J. DAVIS, C. BERRY, A. LEON, H. SCHRYVER, J. KIM, S. MCCULLOCH, J. FRIEDRICH, C. LAITON, J. WONG, O. ZOBEIRI, I. YAVORSKA, E. PETERSON, A. ARCHIPOV, S. OLSEN, P. GROBLEWSKI, S. SESHAMANI, D. FENG, M. GARRETT; Allen Inst., Seattle, WA

Abstract: Neural circuits exhibit a myriad of dynamic activity patterns across numerous cell types to produce adaptative behavior and learning. Specific cell sub-classes (e.g. inhibitory VIP, PV, and SST or excitatory IT and PT) have distinct roles in microcircuit computations and behavior. However, single cell RNA profiling offers more granular definitions of cell types (Tasic et al., 2016; Yao et al., 2021). Very little is known about the functional significance of the hundreds of transcriptomically defined cell types. Combining in vivo optical physiology with post-hoc spatial transcriptomics promises to link circuit function during behavior to patterns of gene expression (Bugeon et al., 2022; Condylis et al., 2022). Here, we present an open-source, cloud-based pipeline for integrating longitudinal 2-photon calcium imaging with lightsheet multiplexed fluorescence in-situ hybridization (mFISH). Our approach employs a blend of community-based tools and custom solutions, with quality control evaluated at each step of data collection and processing. For 2-photon imaging, minimizing spatial drift and reliable cell segmentation aid confident neuron identification across weeks of longitudinal imaging. In lightsheet data analysis, we perform tile stitching, 3D cell-body segmentation, and registration between subsequent rounds of lightsheet imaging and with structural 2-photon images. Aligning ex vivo transcriptomics data with in vivo recordings necessitates non-rigid transformation which is aided by fiducial markers. We evaluated manual and algorithmic approaches for cell co-registration, measuring performance across experimental conditions. The integration of these techniques and our openly available data processing pipeline serve as a resource to support the investigation of cellular-level gene expression patterns of neurons recorded in vivo.

Disclosures: M.J. Davis: None. **C. Berry:** None. **A. Leon:** None. **H. Schryver:** None. **J. Kim:** None. **S. McCulloch:** None. **J. Friedrich:** None. **C. Laiton:** None. **J. Wong:** None. **O. Zobeiri:** None. **I. Yavorska:** None. **E. Peterson:** None. **A. Archipov:** None. **S. Olsen:** None. **P. Groblewski:** None. **S. Seshamani:** None. **D. Feng:** None. **M. Garrett:** None.

Poster

PSTR179. Systems Biology and Multiomics Approaches

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR179.13/VV45

Topic: I.02. Systems Biology and Bioinformatics

Support: NIH Grant U01MH130907

Title: Bridging Function and Transcriptomics: Insights from Visual Responses in the Mouse Visual Cortex during Passive Viewing

Authors: *O. ZOBEIRI[1], H. SCHRYVER[1], C. BERRY[1], V. HAYNES[1], S. MCCULLOCH[1], J. KIM[1], M. DAVIS[1], E. PETERSON[1], L. SUAREZ[1], J. WILKES[1], M. TAORMINA[1], S. CALDEJON[1], A. WILLIFORD[1], J. MILLER[2], J. WATERS[3], S. OLSEN[1], P. A. GROBLEWSKI[3], M. GARRETT[2], A. ARKHIPOV[3];
[1]Allen Inst., Seattle, WA; [2]Allen Inst. for Brain Sci., Seattle, WA; [3]Allen Inst. For Brain Sci., Seattle, WA

Abstract: The powerful functional properties of the brain can be observed through the computations and patterns of activity exhibited by neurons, which include numerous transcriptomically defined cell types within any given brain region. Although there is evidence supporting cell-type-specific functional response to stimuli or changing behavioral state, the extent of these relationships remains uncertain. A key question is whether and how transcriptomically defined cell types make distinct contributions to the underlying mechanisms of broader circuit properties, such as the functional types of neuronal activity. To address this question, we are studying the responses of neurons in the visual cortex. We utilized two-photon calcium imaging to analyze neuron responses in six transgenic mouse lines from a previously published dataset, as well as three additional mouse lines (Gad2, Cux2, Rbp4) from a new dataset. In subsets of Gad2 inhibitory neurons, we employed Hybridization Chain Reaction (HCR) mFISH to determine gene expression, enabling us to discern major hierarchical branches of inhibitory neuron subclasses and further subdivisions within certain cell types. Neuronal responses were recorded while presenting drifting sinusoidal gratings in eight directions and five contrasts to study specific visual selectivity properties in each mouse line. We employed a generalized linear model to investigate the influence of visual (contrast and direction) and non-visual factors (locomotion and pupil diameter) on neuronal responses.Our modeling approach revealed that over 50% of neurons responded to visual and/or non-visual parameters, with a higher prevalence of responsive cells observed in inhibitory mouse lines. Additionally, we observed that neurons encoding the most information about visual motion direction carried significantly more information about either behavioral signals or visual signals. Neurons carrying the least direction information also carried the least information overall. This heterogeneity in functional properties highlights avenues for investigating potential contributions of different cell types to circuit computations underlying the interactions between visual and behavioral signals in visual cortex. Further studies will explore whether the observed heterogeneity corresponds to distinct transcriptomic cell types.

Disclosures: O. Zobeiri: None. **H. Schryver:** None. **C. Berry:** None. **V. Haynes:** None. **S. McCulloch:** None. **J. Kim:** None. **M. Davis:** None. **E. Peterson:** None. **L. Suarez:** None. **J. Wilkes:** None. **M. Taormina:** None. **S. Caldejon:** None. **A. Williford:** None. **J. Miller:** None. **J. Waters:** None. **S. Olsen:** None. **P.A. Groblewski:** None. **M. Garrett:** None. **A. Arkhipov:** None.

Poster

PSTR179. Systems Biology and Multiomics Approaches

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR179.14/VV46

Topic: I.02. Systems Biology and Bioinformatics

Support: NIH Grant U01MH130907

Title: Bridging function and transcriptomics: Quantifying gene expression in the mouse visual cortex

Authors: *H. SCHRYVER, C. BERRY, S. MCCULLOCH, O. ZOBEIRI, J. KIM, M. DAVIS, E. PETERSON, C. BONATTO PAESE, L. SUAREZ, J. WILKES, M. TAORMINA, S. CALDEJON, A. WILLIFORD, J. MILLER, J. WATERS, S. OLSEN, P. GROBLEWSKI, M. GARRETT, A. ARKHIPOV;
Allen Inst., Seattle, WA

Abstract: Understanding the relationship between cell types and circuit properties is crucial for unraveling the brain's functional properties, shaped by neuronal activity patterns and computations that evolve through learning, homeostatic plasticity, and other processes. However, the precise nature of these relationships, particularly at the fine-grained level with a multitude of cell types in any given brain region, remains unclear. Active research is underway to explore the concept of cell types, especially regarding their specific contributions to broader circuit properties and functional types of neuronal activity *in vivo*. We employed multiplexed fluorescence in-situ hybridization (mFISH) to map cells to transcriptomic types in the mouse primary visual cortex (V1). Leveraging Allen Institute single-cell RNAseq data, we designed gene panels that allowed us to resolve inhibitory subclasses and achieve 70% accuracy in mapping clusters within those subclasses using a minimal panel of 15 genes. To capture the expression of different genes for the same cell across multiple imaging rounds, consecutive rounds were registered. We quantified gene expression by examining the luminance levels of pixels within segmented cell masks relative to the local background luminance level, utilizing lightsheet-imaged tissue. By applying a threshold, we determined the binary expression status (positive or negative) of each gene in individual cells. This approach successfully revealed major hierarchical branches of inhibitory neuron types (subclasses) and further subdivisions within specific cell types. We validated our approach by comparing the co-expression patterns of specific genes across our cells with Allen Institute scRNAseq data, as well as other datasets quantifying the relative expression of inhibitory subclasses in mouse V1. Encouragingly, we found similarities across animals and observed consistency when compared to existing datasets. Furthermore, our findings aligned with known patterns of gene expression across different depths of V1. To establish a link between transcriptomic types and functional responses, we registered populations of these cells to those recorded using two-photon calcium imaging *in vivo*. This integration enables us to examine the relationship between transcriptomically defined cells and visual responses and neural activity during behavior. Moving forward, we anticipate that

these methods of quantifying transcriptomically defined cells will shed light on the intricate relationship between cell types and the broader aspects of brain function.

Disclosures: **H. Schryver:** None. **C. Berry:** None. **S. McCulloch:** None. **O. Zobeiri:** None. **J. Kim:** None. **M. Davis:** None. **E. Peterson:** None. **C. Bonatto Paese:** None. **L. Suarez:** None. **J. Wilkes:** None. **M. Taormina:** None. **S. Caldejon:** None. **A. Williford:** None. **J. Miller:** None. **J. Waters:** None. **S. Olsen:** None. **P. Groblewski:** None. **M. Garrett:** None. **A. Arkhipov:** None.

Poster

PSTR179. Systems Biology and Multiomics Approaches

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR179.15/VV47

Topic: I.02. Systems Biology and Bioinformatics

Support: NIH Grant 1U01MH130907-01

Title: Linking functional activity and circuit structure in the mouse primary visual cortex

Authors: ***J. AMAN**[1], O. ZOBEIRI[1], C. KING[1], M. A. BUICE[1], N. DA COSTA[2], F. COLLMAN[2], C. SCHNEIDER-MIZELL[2], A. ARKHIPOV[2];
[1]Allen Inst., Seattle, WA; [2]Allen Inst. For Brain Sci., Seattle, WA

Abstract: Cerebral cortex contains a diversity of cell types, each with unique anatomical, molecular and functional properties. A longstanding question is whether neurons with distinct morphological or connectivity properties have distinct physiological activity. Here, we characterize the responses of V1 excitatory neurons to visual stimuli and locomotion, and ask whether neurons with shared response properties have similar morphologies and connectivity. We use an Allen Institute dataset in which calcium imaging data were co-registered with electron microscopy (EM) data of the same volume. Activity of excitatory neurons in an 800x800 micron column of V1 spanning all cortical layers was recorded using 2-photon (2p) calcium imaging. During imaging sessions, the mouse freely ran on a wheel and was presented with a variety of visual stimuli, including drifting gratings, sparse noise, natural images, and natural movies. We used a generalized linear model (GLM) to predict trial-averaged calcium responses from visual stimuli and the mouse's behavioral state. This model performs well for a subset of cells (~15%). For these cells, the mouse's running speed and pupil diameter are poor predictors of neural responses. Instead, we find that visual stimulus features are predictive of neuronal activity. We identify clusters of excitatory cells that have shared responsiveness for drifting grating and natural movie stimuli. For cells that are co-registered to the EM volume, we compare visual cluster membership with previously identified morphological cell types in layer 4 and layer 2/3. We further explore whether a cell's input connectivity predicts visual cluster membership.

Disclosures: **J. Aman:** None. **O. Zobeiri:** None. **C. King:** None. **M.A. Buice:** None. **N. da Costa:** None. **F. Collman:** None. **C. Schneider-Mizell:** None. **A. Arkhipov:** None.

Poster

PSTR179. Systems Biology and Multiomics Approaches

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR179.16/VV48

Topic: I.02. Systems Biology and Bioinformatics

Support: 1U19NS123714

Title: Targeting thalamus cell populations with enhancer-driven AAV vectors

Authors: *M. J. HOOPER[1], S. YAO[1], B. R. LONG[1], S. HUFF[1], J. ROTH[1], B. TASIC[1], N. DONADIO[1], M. GABITTO[1], T. CHARTRAND[1], F. HAESELEER[1], J. T. TING[1], B. P. LEVI[1], Y. BEN-SIMON[1], M. TAORMINA[1], C. VAN VELTHOVEN[1], M. KUNST[1], A. OSTER[1], S. WAY[1], B. THYAGARAJAN[1], E. LEIN[1], J. NGAI[2], H. ZENG[1], K. SVOBODA[1];
[1]Allen Inst., Seattle, WA; [2]NIH, Bethesda, MD

Abstract: Purpose:
The thalamus functions as a central relay that takes input from sensory systems and subcortical brain regions, and gives output to the cerebral cortex. The development of tools for selective access to different thalamic cell populations will enable studies of their connectivity and function. The goal of this work is to develop recombinant enhancer-driven adeno-associated viral (AAV) vectors to target thalamic cell populations.
Methods:
Single-cell multiome data allows simultaneous examination of RNA and accessible chromatin in single cells. Using these data, we identified putative population-specific enhancer elements. Putative enhancer elements were used for generation of enhancer-driven SYFP2 AAV vectors. Enhancer AAVs were injected retroorbitally in mice 3 weeks prior to brain collection for histology. SYFP2 expression in the target population was evaluated based on the spatial location of the equivalent cell populations in Multiplexed error-robust fluorescence in situ hybridization (MERFISH) data. Further validation of cell population was done using serial two photon tomography (STPT) and single-cell RNA sequencing.
Results:
81 AAV vectors designed to target thalamus subpopulations were generated and tested. 49% of vectors targeting glutamatergic populations showed signal in the target population. STPT experiments showed that enhancer AAVs could induce SYFP2 expression in spatial patterns that were consistent with the distribution of the target population in MERFISH data.
Conclusions:
We developed a set of enhancer AAV tools based on single cell chromatin accessibility data and validated them using histology. The spatial distribution of target cell populations in MERFISH data often were consistent with SYFP2 expression in STPT experiments. Future functional studies of thalamus cell types will make use of Cre and Flp vectors generated using these enhancer sequences. Effort is ongoing to expand the breadth of our suite of tools at higher cell type resolution.

Disclosures: **M.J. Hooper:** None. **S. Yao:** None. **B.R. Long:** None. **S. Huff:** None. **J. Roth:** None. **B. Tasic:** None. **N. Donadio:** None. **M. Gabitto:** None. **T. Chartrand:** None. **F. Haeseleer:** None. **J.T. Ting:** None. **B.P. Levi:** None. **Y. Ben-Simon:** None. **M. Taormina:** None. **C. van Velthoven:** None. **M. Kunst:** None. **A. Oster:** None. **S. Way:** None. **B. Thyagarajan:** None. **E. Lein:** None. **J. Ngai:** None. **H. Zeng:** None. **K. Svoboda:** None.

Poster

PSTR179. Systems Biology and Multiomics Approaches

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR179.17/VV49

Topic: I.02. Systems Biology and Bioinformatics

Title: Spatial proteomic analysis of alzheimer's disease human brain using multiplexed imaging

Authors: *A. BOSE[1], L. ARVIDSON[2], R. W. CHO[2], S. SINGH[2], G. SPANG[2], M. J. SMITH[1], V. AGRAWAL[1];
[1]Leica Microsystems, Waltham, MA; [2]Cell Signaling Technol., Danvers, MA

Abstract: Alzheimer's disease (AD) is a genetic and sporadic neurodegenerative disease and a common cause of cognitive impairment acquired in midlife and late life. AD is pathologically defined by the presence of β-amyloid-containing plaques and phosphorylated tau containing neurofibrillary tangles. Effective therapies for the treatment and/or prevention of AD are lacking. The understanding of how synaptic degeneration occurs and the contribution of glial and peripheral cells to disease progression is essential for the development of new therapies. The examination of the histopathological features of AD may reveal cellular relationships that contribute to disease etiology leading to potential novel therapeutic strategies. The Cell DIVE Multiplexed Imaging Solution, in combination with IF/IHC-validated antibodies from Cell Signaling Technology (CST), can be used to computationally examine synaptic processes and spatially and molecularly define cells, such as glia and neurons, surrounding pathological hallmarks in AD. Segmentation and clustering analysis can identify spatially co-localized populations of cells, including subpopulations of microglia defined by specific disease-associated microglia markers. CST's broad portfolio of validated antibodies enables the detection of altered synaptic processes and cell-type populations in the context of human disease tissue. Here we demonstrate multiplexed Cell DIVE imaging using a novel CST panel to probe AD brain. We examine the protein landscape in diseased tissue in the context of Amyloid-β (Aβ) and tau expression. The ability of cell type specific markers, combined with multiplexed tissue imaging will provide a new approach for the neuroscience research community to understand spatial heterogeneity of the human brain and their contribution to disease.

Disclosures: **A. Bose:** A. Employment/Salary (full or part-time); Leica Microsystems. **L. Arvidson:** A. Employment/Salary (full or part-time); Cell Signaling Technology. **R.W. Cho:** A. Employment/Salary (full or part-time); Cell Signaling Technology. **S. Singh:** A.

Employment/Salary (full or part-time); Cell Signaling Technology. **G. Spang:** A. Employment/Salary (full or part-time); Cell Signaling Technology. **M.J. Smith:** A. Employment/Salary (full or part-time); Leica Microsystems. **V. Agrawal:** A. Employment/Salary (full or part-time); Leica Microsystems.

Poster

PSTR179. Systems Biology and Multiomics Approaches

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR179.18/VV50

Topic: I.02. Systems Biology and Bioinformatics

Support: BRAIN Initiative NIH/NIMH U01MH117079
BRAIN Initiative NIH/NIMH RF1MH128888
CHDI

Title: Large-scale Mapping of Dendritic Morphology in Striatal Medium Spiny Neurons of Wildtype and Huntington's Disease Mice

Authors: *C. PARK[1,2,4], C. CHOI[1,2,4], M. ZHU[1,3], K. MARRETT[1,5], M. YAN[1,2,4], G. MAGAT[1,2,4], M. AKRAM[1,2,4], A. BANNECKE[1,6], S. ZHANG[1,2,4], R. VACA[1,2,4], Y. KIM[7], G. ASCOLI[8], J. CONG[1,5], D. TWARD[1,6], H. DONG[1,3], X. YANG[1,2,4];
[1]UCLA, Los Angeles, CA; [2]Dept. of Psychiatry and Biobehavioral Sci., [3]UCLA Brain Res. and Artificial Intelligence Nexus, Dept. of Neurobio., David Geffen Sch. of Med. at UCLA, Los Angeles, CA; [4]Ctr. for Neurobehavioral Genetics, Jane and Terry Semel Inst. for Neurosci., [5]Dept. of Computer Sci., [6]Dept. of Computat. Med. and Neurology, Ahmanson-Lovelace Brain Mapping Ctr., Univ. of California at Los Angeles, Los Angeles, CA; [7]Dept. of Neural and Behavioral Sci., Penn State Univ., Hershey, PA; [8]Ctr. for Neural Informatics, Structures, & Plasticity and Bioengineering Dept., George Mason Univ., Fairfax, VA

Abstract: Despite the critical role played by dendrites in understanding the mammalian neuronal biology including connectivity and brain diseases, there is currently a lack of a systems-biology approach to analyze the intact dendritic arbors of genetically-defined neurons in the brain, ranging from labeling and imaging to reconstruction and registration onto reference brain atlas and quantitative analysis. Here we provide a a generalizable and scalable pipeline from genetic sparse and bright labeling of the complete morphology of hundreds or more single-neurons with MORF3 mice (Veldman et al. Neuron, 2020, PMID: 32795398), tissue-clearing and 3D-imaging (e.g., light-sheet) of thick brain sections or intact brain hemispheres, customized multiscale reference map registration, semi-automated single-neuron reconstruction, integrative morphometric analyses, and brainwide single-neuron visualization. With this streamlined pipeline, we developed a reference medium spiny neuron (MSN) single-neuron dendritic feature/anatomical map for over 3500 striatal D1- and D2-MSNs in the brains of P56 C57BL/6J mice, defining both MSN cell-type- and striatal subregion-specific morphometric features. Moreover, we implemented this approach to analyze the MORF3-labeled MSNs in the Q140

knock-in mouse model of Huntington's disease and control mouse brains at 12-month of age. Our study reveals novel MSN dendritic morphological changes in a specific striatal subregion of the HD mouse model, highlighting the importance of brainwide single neuron dendritome mapping as an unbiased approach to characterize normal neuronal morphological variations and more precisely identify pathological changes at single-neuron resolution.

Disclosures: **C. Park:** None. **C. Choi:** None. **M. Zhu:** None. **K. Marrett:** None. **M. Yan:** None. **G. Magat:** None. **M. Akram:** None. **A. Bannecke:** None. **S. Zhang:** None. **R. Vaca:** None. **Y. Kim:** None. **G. Ascoli:** None. **J. Cong:** None. **D. Tward:** None. **H. Dong:** None. **X. Yang:** None.

Poster

PSTR179. Systems Biology and Multiomics Approaches

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR179.19/VV51

Topic: A.08. Development of Neural Systems

Support: Zhejiang Province Natural Science Foundation (LR20C070002)

Title: Performing de novo reconstruction of neuron networks in asexual planarian using 3D tissue imaging by tiling light sheet microscopy

Authors: *J. LU, H. XU, L. GAO, K. LEI;
Westlake Univ., Hangzhou, China

Abstract: Damage to the central nervous system in adult mammals is irreversible. The freshwater planarian exhibits an ancient pair of central nervous systems and the ultimate ability of regeneration. Spatial information of neuron cells and regulation factors during regeneration is largely unknown. Here we present a 3D tissue reconstruction method to investigate neuron type diversity and development at single-cell level with the labelling of various neuron types, including cholinergic, GABAergic, octopaminergic, dopaminergic and serotonergic neurons. We modified the expansion microscopy technique based on tissue clearing to achieve nanoscale imaging of planarians at under 100 nm resolution with up to 5* linear expansion. We validated and demonstrated the 3D tissue reconstruction method with over 400 wide-type planarians from all stages of regeneration and homeostasis, with a pipeline to count and locate planarian single neuron and body cells. As a planarian's body size increases, all neuron subtypes grow alongside the body cells until a threshold is reached, suggesting a concordant with planarian fission. To examine the interaction between muscles and neurons during the regeneration process, we examined the structure, location, and regeneration of muscle fibers, as well as their connection to cholinergic neurons and glial cells labeled with Estrella. We found out that the ventral muscle fibers located in the inner epidermal layer of the planarian body locate closely with cholinergic neurons and glial branches. These findings provide evidence that muscle fiber could act as a scaffold for neuron targeting. Moreover, we proposed the 3D tissue reconstruction method to be

a versatile tool for super-resolution and large-scale imaging for planarians and other small animals.

Disclosures: **J. Lu:** None. **H. Xu:** None. **L. Gao:** None. **K. Lei:** None.

Poster

PSTR180. Biomarkers for Neurodegenerative Diseases

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR180.01/VV52

Topic: I.05. Biomarker, Drug Discovery, and Experimental Therapeutics

Title: The importance of neurofilament light chain (NF-L) biomarker in 5XFAD Alzheimer's disease and SOD1-G93A amyotrophic lateral sclerosis animal disease models

Authors: *S.-Y. NA, Y. KIM, S. LEE, Y. KANG, Y. JUNG, M. LEE, J. LEE, P. SWEENEY, K. PARK, L. PARK;
Naason Sci. Inc., Cheongju-si, Chungbuk, Korea, Republic of

Abstract: The identification of neurofilament subunits in cerebrospinal fluid (CSF) and blood has emerged as a prevalent method for evaluating axonal compromise, providing valuable insights into the extent of axonal damage in neurodegenerative diseases. Neurofilament light chain (NF-L), along with other major neurofilament subunits (NF-M, NF-H, and α-internexin), forms heteropolymers that assemble into 10nm neurofilaments primarily found in neurons, especially in large projection axons. Among these subunits, NF-L has gained prominence as a clinical biomarker. The measurement of NF-L levels in CSF and blood holds significant potential for early detection, prognosis, and monitoring of neurodegenerative disorders. Conditions such as Alzheimer's disease (AD) and amyotrophic lateral sclerosis (ALS) are characterized by the accumulation of abnormal proteins, including neurofilament subunits, which ultimately lead to neuronal degeneration and cognitive decline. Detecting NF-L in CSF and blood enables early intervention and the formulation of potential treatment strategies. Moreover, NF-L serves as an important biomarker in animal models of AD and ALS. In this study, we aimed to investigate the changes in NF-L levels that may correlate to disease progression and aging in animal models of 5XFAD AD and SOD1-G93A ALS mice. Specifically, we examined NF-L concentration in 5xFAD mice aged 3 to 15 months and SOD1-G93A mice aged 11 to 21 weeks. In cognitive tests such as the Y-maze and Morris water maze, the spontaneous alteration behavior of 5xFAD mice was shorter than that of the control group after 9 months of age. Neurological and disease scores of SOD1-G93A mice exhibited age-dependent increases from 11 to 21 weeks. Our findings revealed that NF-L concentration in both 5xFAD and SOD1-G93A mice increased with age and disease progression. These results indicate that changes in NF-L levels may serve as an early indicator of disease progression before the onset of cognitive or motor impairments. Monitoring NF-L levels in CSF and blood, therefore, holds promise for providing valuable insights into neurodegenerative diseases and potentially aiding in the development of therapeutic interventions.

Disclosures: **S. Na:** None. **Y. Kim:** None. **S. Lee:** None. **Y. Kang:** None. **Y. Jung:** None. **M. Lee:** None. **J. Lee:** None. **P. Sweeney:** None. **K. Park:** None. **L. Park:** None.

Poster

PSTR180. Biomarkers for Neurodegenerative Diseases

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR180.02/VV53

Topic: I.05. Biomarker, Drug Discovery, and Experimental Therapeutics

Support: NIH Grant U01NS114144

Title: Analytical Validation of Ultrasensitive Neuroinjury Multiplex Assay Panel

Authors: *C. DEMOS, D. ROMERO, N. PADMANABHAN, R. COHEN, J. BROWN, C. CAMPBELL, M. STENGELIN, A. MATHEW, G. SIGAL, J. WOHLSTADTER;
Meso Scale Discovery, Rockville, MD

Abstract: Three blood-based biomarkers of neuroinjury—glial fibrillary acidic protein (GFAP), neurofilament light (Nf-L), and total Tau (tTau)—have emerged as promising biomarkers of neurological disorders and neuroinjuries such as hypoxic-ischemic encephalopathy (HIE), traumatic brain injury (TBI) and Alzheimer's disease (AD). The low levels of GFAP, Nf-L, and tTau in serum and plasma require highly sensitive assays to detect these important biomarkers. Here, we report on an ultrasensitive, electrochemiluminescence-based, multiplexed immunoassay for GFAP, Nf-L, and tTau that has been analytically validated, providing a new tool for assessing neuroinjury biomarkers. The MSD S-PLEX Neurology Panel 1 kit uses ultrasensitive S-PLEX assay technology to simultaneously measure GFAP, Nf-L, and tTau in a 96-well plate format using standard liquid-handling techniques. Analytical validation was carried out in a series of studies based in part on Clinical and Laboratory Standards Institute guidelines EP05-A3, EP06-Ed2, EP07-Ed3, EP17-A2, EP25-A, and EP28-A3c to evaluate precision, dilution linearity, interference screening, detection capability, stability, spike recovery, and cross reactivity. The assay requires 5 µL of a sample for each replicate. The time to result is less than 6 hours and the quantifiable range of the assay is 2.4-4,230 pg/mL for GFAP, 7.65-10,000 pg/mL for Nf-L, and 0.34-735 pg/mL for tTau. A set of 15 common potentially interfering substances were screened in serum and plasma, and none showed interference exceeding 18% difference in measurement compared to non-spiked samples. Within-laboratory precision was <11% CV for GFAP and Nf-L and <19% CV for tTau for samples spanning the reportable range. Dilution up to 256-fold recovered within 80-120% of the expected value. MSD has developed and analytically validated a highly sensitive, multiplexed immunoassay for quantifying GFAP, Nf-L, and tTau in plasma and serum. This assay is sufficiently sensitive to detect these analytes in normal serum and plasma and has a large dynamic range to accommodate elevated levels found in some neurological disorders.

Disclosures: **C. Demos:** A. Employment/Salary (full or part-time); Meso Scale Diagnostics, LLC. **D. Romero:** A. Employment/Salary (full or part-time); Meso Scale Diagnostics, LLC. **N. Padmanabhan:** A. Employment/Salary (full or part-time); Meso Scale Diagnostics, LLC. **R. Cohen:** A. Employment/Salary (full or part-time); Meso Scale Diagnostics, LLC. **J. Brown:** A. Employment/Salary (full or part-time); Meso Scale Discovery. **C. Campbell:** A. Employment/Salary (full or part-time); Meso Scale Diagnostics, LLC. **M. Stengelin:** A. Employment/Salary (full or part-time); Meso Scale Diagnostics, LLC. **A. Mathew:** A. Employment/Salary (full or part-time); Meso Scale Diagnostics, LLC. **G. Sigal:** A. Employment/Salary (full or part-time); Meso Scale Diagnostics, LLC. **J. Wohlstadter:** E. Ownership Interest (stock, stock options, royalty, receipt of intellectual property rights/patent holder, excluding diversified mutual funds); Meso Scale Diagnostics, LLC.

Poster

PSTR180. Biomarkers for Neurodegenerative Diseases

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR180.03/VV54

Topic: I.05. Biomarker, Drug Discovery, and Experimental Therapeutics

Support: University of Manitoba
Health Science Centre Foundation
NSERC

Title: Automated FDG-PET Reading for Differential Diagnosis of Dementia Spectrum Disorders

Authors: *N. JUNANKAR[1,2], J. PERRON[3,1], J. KO[3,2,1];
[2]Human Anat. and Cell Sci., [3]Grad. Program in Biomed. Engin., [1]Univ. of Manitoba, Winnipeg, MB, Canada

Abstract: Automated FDG-PET Reading for Differential Diagnosis of Dementia Spectrum Disorders

Authors*N. Junankar[1,2], J. Perron[2,3], J. H. Ko[1,2,31] Department of Human Anatomy and Cell Science, Max Rady College of Medicine, Rady Faculty of Health Sciences, University of Manitoba, Winnipeg, MB R3E 0W2, Canada, [2]PrairieNeuro Brain Research Centre, Kleysen Institute for Advanced Medicine, Health Sciences Centre, Winnipeg, MB R3E 3J7, Canada; [3]Graduate Program in Biomedical Engineering, Price Faculty of Engineering, University of Manitoba, Winnipeg, MB R3T 2N2, Canada.

DisclosuresN. Junankar: None**. J. Perron**: None. **J. H. Ko**: None.Neurodegenerative dementia disorders are characterized by a progressive decline in cognitive functions which impact daily life, however these consist of a broad spectrum of diseases each with unique pathology and diagnostics. The clinical standard for differential diagnosis of dementia spectrum disorders (DSD) is brain fluorodeoxyglucose positron emission tomography (FDG-PET) because DSD may be identified by distinct spatial patterns of resting cerebral glucose metabolism. It is a common practice for physicians to interpret these investigations based on subjective impressions

of tracer uptake throughout the brain. In this study, we present an automated approach for the differential diagnosis of DSDs with FDG-PET. 212 patients underwent brain FDG-PET for dementia differential diagnoses: Alzheimer's disease (AD), behavioral-variant frontotemporal dementia (bvFTD), dementia with Lewy body (DLB) and primary progressive aphasia (PPA). 54 patients were diagnosed with dementia (AD = 25, DLB = 17, bvFTD = 6, PPA = 6), 26 had mild cognitive impairment (MCI), 9 were cognitively normal, 12 had non-cognitive conditions and 111 patients lacked follow-up. FDG-PET images were preprocessed in SPM12, underwent SSM, and used to train a KNN. A novel method of data augmentation was used due to the small sample size. Leave-one-out cross-validation on the KNN resulted in 25% and 96% accuracy without and with data augmentation respectively. Results show that data augmentation combined with SSM-based feature selection and classification with a KNN classifier is an effective means of automating readings of FDG-PET images for the differential diagnosis of neurodegenerative DSD.

Disclosures: **N. Junankar:** None. **J. Perron:** None. **J. Ko:** None.

Poster

PSTR180. Biomarkers for Neurodegenerative Diseases

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR180.04/VV55

Topic: I.05. Biomarker, Drug Discovery, and Experimental Therapeutics

Title: High content phenotypic screening platform ATRIVIEW® for compounds that induce neurogenesis and neuroprotection mechanisms in the neurodegenerative environment

Authors: *M.-Y. KIM[1], J.-H. HWANG[1], S. LEE[1], Y. SHIM[1], S. HAN[1,2];
[1]Genuv Inc., Seoul, Korea, Republic of; [2]Genuv US Subsidiary, Cambridge, MA

Abstract: Neurodegenerative disease is characterized by loss of structure or function of neurons, including neuronal death, among many other unknown factors. Although neurotoxic factors were thought to be the target for neurodegenerative disease treatment, since these patients have already undergone significant neuronal damage, such disease-modifying therapies have had limited success. As a new therapeutic approach for treating neurodegenerative diseases, we have identified the importance of neuro-regeneration and neuroprotection by mechanistic studies: those leading to pathophysiological rescues by inducing neurogenesis/neuroprotection with SNR1611(trametinib, active pharmaceutical ingredient of Mekinist®) in Alzheimer's disease (AD) and amyotrophic lateral sclerosis (ALS). The studies and findings were based upon a high-content screening (HCS) system (ATRIVIEW®-HCS) for identifying therapeutic compounds which induce these mechanisms. ATRIVIEW®-HCS is a phenotypic and biomarker-based drug screening system using neural stem cells derived from neurodegenerative disease model mice. It analyzes changes in morphology and neurogenesis/neuroprotection biomarker expression simultaneously. We screened almost 11,000 chemicals using the ATRIVIEW®-HCS platform and selected hits with neuro-regenerative/neuroprotective effects using SNR1611 as a positive

control. Using high-depth resolution confocal images, we successfully established ATRIVIEW®-AI (artificial intelligence) which automates analysis of a large number of confocal images by incorporating deep learning technology into the ATRIVIEW®-HCS platform. The technology classifies cells by type such as differentiated, toxic, or non-changed cells, etc. and recognizes different regions of individual cells. In addition, the expression levels of the biomarkers of choice as well as morphological feature parameters are automatically calculated. They are visualized on top of the 96-well plate images to make it easier for users to analyze the multi-data output to determine potential hits intuitively. Thus, the rapid and advanced ATRIVIEW®-AI platform increases screening efficiency by dramatically reducing the resources and time required for image analysis while enhancing acuity. Furthermore, it allows evaluation of biomarkers underlying the processes of neurogenesis/neuroprotection in the primary cell culture, increasing the potential of identifying drug candidates for the treatment of neurodegenerative disease.

Disclosures: **M. Kim:** A. Employment/Salary (full or part-time); Genuv Inc.. E. Ownership Interest (stock, stock options, royalty, receipt of intellectual property rights/patent holder, excluding diversified mutual funds); Genuv Inc. **J. Hwang:** A. Employment/Salary (full or part-time); Genuv Inc. **S. Lee:** A. Employment/Salary (full or part-time); Genuv Inc. **Y. Shim:** A. Employment/Salary (full or part-time); Genuv Inc. **S. Han:** A. Employment/Salary (full or part-time); Genuv Inc.. E. Ownership Interest (stock, stock options, royalty, receipt of intellectual property rights/patent holder, excluding diversified mutual funds); Genuv Inc.

Poster

PSTR180. Biomarkers for Neurodegenerative Diseases

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR180.05/VV56

Topic: I.05. Biomarker, Drug Discovery, and Experimental Therapeutics

Title: Ultrasensitive detection of total and phosphorylated Tau protein in human blood using Single Molecule Counting (SMC)Technology

Authors: *F. WIESE, M. LIPPOLD, J. FISKE, R. GITAN;
Milliporesigma, St Louis, MO

Abstract: Tau, the microtubule associated protein, is misfolded and forms aggregates in the brains of individuals with Alzheimer's disease. Detection of increased levels of total tau protein, as well as phosphorylated tau protein in cerebrospinal fluid (CSF) is indicative of Alzheimer's Disease. The increased levels of Tau protein present in Alzheimer's Disease is also detectable in blood, though highly specific and sensitive immunoassays are needed for this detection. We have developed ultra-high sensitivity immunoassays on the Single Molecule Counting (SMC®) platform to allow detection of total Tau and phosphorylated Tau. These four assays allow detection of total, pT181, pT217 and pT231 Tau proteins in human serum and plasma at levels down to less than 1 pg/mL. Statistically significant differential expression of total and phosphorylated Tau protein is observed when sample concentrations are compared between

normal and Alzheimer's disease samples. These assays represent a powerful tool for Alzheimer's disease research, enabling the detection and quantification of blood borne Tau protein.

Disclosures: F. Wiese: A. Employment/Salary (full or part-time); MilliporeSigma. **M. Lippold:** A. Employment/Salary (full or part-time); MilliporeSigma. **J. Fiske:** A. Employment/Salary (full or part-time); MilliporeSigma. **R. Gitan:** A. Employment/Salary (full or part-time); MilliporeSigma.

Poster

PSTR180. Biomarkers for Neurodegenerative Diseases

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR180.06

Topic: I.05. Biomarker, Drug Discovery, and Experimental Therapeutics

Title: Development and validation of an α-Synuclein aggregate ELISA for analyzing human and transgenic model samples.

Authors: *A. NIKOLENKO, C. EVANS, W.-C. LIAO, B. SUN, J. NI;
BioLegend, San Diego, CA

Abstract: The ELISA has been one of the most authoritative research tools for various biomarkers detection and measurement. It is especially beneficial for studying aggregated proteins linked to neurodegenerative diseases, such as amyloid β (Alzheimer's disease, AD) and α-synuclein (Parkinson's disease, PD). BioLegend - in collaboration with the Michael J. Fox Foundation - has recently developed a new ELISA kit for PD research. This kit can be used to analyze various sample types, such as human brain lysates and cerebrospinal fluid (CSF) samples, as well as lysates from transgenic mouse and fruit fly model systems. In this work, we describe the development and validation of an ELISA assay, which utilizes α-synuclein aggregate-specific antibodies and an aggregated protein (each produced in-house). These reagents were thoroughly optimized to develop a highly robust and reliable ELISA assay that can consistently measure levels of α-synuclein aggregates in various biological sample types. Furthermore, the kit was validated to accurately measure α-synuclein aggregate concentrations in human A53T-α-synuclein-expressing mouse brain lysates and human α-synuclein transgenic fruit fly head lysates. We found that in the mouse brain lysates, the ratio of aggregate-to-total α-synuclein was higher for lysates mimicking PD (compared to control lysates). The observed result is supported by published data, and it demonstrates the practicality of the LEGEND MAX™ α-Synuclein Aggregate ELISA as a valuable addition to the library of the available PD research tools for α-synuclein aggregate detection.

Disclosures: A. Nikolenko: None. **C. Evans:** None. **W. Liao:** None. **B. Sun:** None. **J. Ni:** None.

Poster

PSTR180. Biomarkers for Neurodegenerative Diseases

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR180.07/VV57

Topic: I.05. Biomarker, Drug Discovery, and Experimental Therapeutics

Title: Wearable inertial measurement units for movement classification in ice dance patterns using root mean square distance as a metric

Authors: *A. G. YING[1], S. G. YING[1], Y. S. TARUI[1], A. V. JOYCE[1], C. S. DUGAN[1], A. T. JOYCE[1], V. A. YE[1], M. E. DUGAN[1], N. S. YING[1], C. F. JOYCE[1], D. S. DUGAN[1], S. PAN[1], M. G. BRAMANTE[1], E. T. GEMINIANI[2], S. H. YING[1];
[1]Deeper Edge, Cambridge, MA; [2]Boston Children's Hosp., Boston, MA

Abstract: Background: Clinical management of neurological movement disorders would benefit from having a quantitative method to measure and rate movements. This proof-of-concept pilot study uses ice dance as a model system for digital movement assessment because ice dance movements are standardized, with a grading system based on precision of execution of a predefined pattern. Our previous work demonstrated the feasibility of using wearable inertial measurement units (IMU's) to measure and classify basic ice dance movements, using Pearson's product-moment correlation coefficient (PMCC) to compare a step in question to a template step. Here we test the hypothesis that classification can be improved by preserving temporal relationships using root mean square distance (RMSD) as the similarity metric instead of PMCC.
Methods: Noitom Perception Neuron IMUs were used to record ice skaters performing the Dutch Waltz, a standardized ice dance pattern consisting of eight steps. The data was grouped and visualized according to the known classification of each step. A template for each ice dance step were created using a numerical average of the data. Because there were clear morphological differences between skaters, templates were individualized to each skater. Each step was classified according to either the lowest RMSD or the highest PMCC; overall efficacy was based on the percentage of steps that were correctly classified.
Results: Two skaters each performed six patterns of the Dutch Waltz, for a total of 48 steps from each skater. Each step was compared to a set of template steps for classification. When using RMSD as a similarity metric, classification accuracy was 94-100%. When using PMCC as a similarity metric, classification accuracy was 81-85%. To assess the statistical significance of these differences, McNemar's test for paired proportions was conducted. The resultant chi-square test statistic was 8.47, indicating a significant difference in the proportions of correct classifications between the two metrics.
Conclusion: Our results demonstrate the feasibility of the collection of data from IMUs during an ice dance pattern dance and the utility of a simple algorithm to classify steps according to an individualized template. Chi-squared analysis showed that using the root mean square distance as a distance metric produced a higher percent classification accuracy as compared to using the Pearson product-moment correlation coefficient. To demonstrate robustness of the algorithm, it is necessary to collect more samples. A larger training set could be useful to develop a strategy for classification across a wider range of individuals, over a wider range of movements.

Disclosures: **A.G. Ying:** None. **S.G. Ying:** None. **Y.S. Tarui:** None. **A.V. Joyce:** None. **C.S. Dugan:** None. **A.T. Joyce:** None. **V.A. Ye:** None. **M.E. Dugan:** None. **N.S. Ying:** None. **C.F. Joyce:** None. **D.S. Dugan:** None. **S. Pan:** None. **M.G. Bramante:** None. **E.T. Geminiani:** None. **S.H. Ying:** None.

Poster

PSTR180. Biomarkers for Neurodegenerative Diseases

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR180.08/VV58

Topic: I.05. Biomarker, Drug Discovery, and Experimental Therapeutics

Support: NIA Grant R01 #AG050598
Sorrell Chesin Research Award
Center for Undergraduate Research and Creative Engagement (CURCE), University at Albany

Title: Relationship between BDNF concentrations in hippocampal tissue and blood sera of Sprague Dawley rats

Authors: *S. E. LEE[1], S. DOUGLASS[2], E. C. MCNAY[3];
[1]State Univ. of New York, Univ. of Albany, Albany, NY; [2]State Univ. of New York At Albany, Albany, NY; [3]Behavioural Neurosci., Univ. At Albany, Albany, NY

Abstract: Brain-derived neurotrophic factor (BDNF) is necessary for neuronal survival, synaptic plasticity, and neurogenesis. Recent studies have shown that dysregulation of BDNF contributes to the development of several neuropsychiatric disorders including major depressive disorder, schizophrenia, and dementia; reduced BDNF correlates with an increase in neuronal atrophy, most prominently in the hippocampus. Further, there is decreased BDNF expression in serum, hippocampi, and cortical tissue from post-mortem suicidal and depressive patients. Thus, BDNF has been identified as a potential biomarker for neurological dysfunction. Routine direct measurement of brain BDNF is implausible; hence, in this study we will examine the relationship between blood and hippocampal BDNF levels in rats, to determine whether peripheral BDNF measures can serve as a proxy for central levels as has been suggested by prior work. We hypothesize that there will be a positive correlation between hippocampal and blood sera BDNF expression in adult Sprague Dawley rats, measured using ELISA, which will support the use of BDNF as a biomarker for neural health.

Disclosures: **S.E. Lee:** None. **S. Douglass:** None. **E.C. McNay:** None.

Poster

PSTR180. Biomarkers for Neurodegenerative Diseases

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR180.09/VV59

Topic: I.05. Biomarker, Drug Discovery, and Experimental Therapeutics

Title: Validation of Novel Ultrasensitive Assays for Detection of Phosphorylated Tau (pT181 and pT217) in Human Plasma, Serum, and Cerebrospinal Fluid

Authors: B. OTIENO[1], Q. NING[1], C. ISLAM[1], L. CHACKO[1], F. MINOOEI[1], R. COHEN[1], *J. RANDALL[2], S. B. HARKINS[1], A. MATHEW[1], L. QUINTERO[1], J. N. WOHLSTADTER[1]; [1]Meso Scale Diagnostics, LLC. (MSD), Rockville, MD, USA, Rockville, MD; [2]Meso Scale Diagnostics, LLC. (MSD), Rockville, MD, USA, Westford, MA

Abstract: Aggregation of phosphorylated tau (pTau) into neurofibrillary tangles is closely associated with Alzheimer's disease (AD). Hyper-phosphorylated tau in the brain has been imaged by classical biochemical techniques like immunohistochemical staining, but *in vivo* histological investigations are not possible. Measurement of circulating levels of tau phosphorylated at unique sites holds promise for early AD detection and for guiding potential treatment, as well as for the selection, classification, and monitoring of patients in clinical trials. Moreover, blood-based biomarker detection assays can simplify patient screening further by removing the need for an invasive spinal tap, which is necessary for collecting cerebrospinal fluid (CSF). An accurate, reliable, and ultra-sensitive, blood-based pTau assay should significantly benefit AD research and fuel new understandings in pre-clinical research and screening.Here, we describe the validation of ultrasensitive pTau (pT181 and pT217) biomarker assays in human plasma, serum, and CSF. Electrochemiluminescent (ECL) assays were performed on the Meso Scale Discovery S-PLEX platform using 96-well plates, different capture antibodies specific to each phosphorylation site of tau, and a common total tau detection antibody. Validation included assessment of intra- and inter-assay accuracy, precision, specificity, dilution linearity, selectivity, and stability. Both assays demonstrate high sensitivity with limits of detection in the femtogram/mL range. The specificity of the assays was confirmed using a non-phosphorylated tau protein and tau proteins lacking the phosphorylation sites. All parameters evaluated met the predefined acceptance criteria per the FDA guidelines for analytical method validation. Matched healthy control (N = 15) and AD patient (N = 15) samples were tested. pTau was detectable in the three matrices, with higher levels of pT181 and pT217 observed in AD patients than in healthy controls. The assays allow for accurate, precise, and ultrasensitive detection of pT181 and pT217 in blood, and they can be used to support research regarding implementation of biomarker assays for AD and monitoring of disease progression and treatment.

Disclosures: **B. Otieno:** A. Employment/Salary (full or part-time); Meso Scale Diagnostics, LLC. (MSD), Rockville, MD, USA. **Q. Ning:** A. Employment/Salary (full or part-time); Meso Scale Diagnostics, LLC. (MSD), Rockville, MD, USA. **C. Islam:** A. Employment/Salary (full or part-time); Meso Scale Diagnostics, LLC. (MSD), Rockville, MD, USA. **L. Chacko:** A. Employment/Salary (full or part-time); Meso Scale Diagnostics, LLC. (MSD), Rockville, MD, USA. **F. Minooei:** A. Employment/Salary (full or part-time); Meso Scale Diagnostics, LLC. (MSD), Rockville, MD, USA. **R. Cohen:** A. Employment/Salary (full or part-time); Meso Scale Diagnostics, LLC. (MSD), Rockville, MD, USA. **J. Randall:** A. Employment/Salary (full or

part-time); Meso Scale Diagnostics, LLC. (MSD), Rockville, MD, USA. **S.B. Harkins:** A. Employment/Salary (full or part-time); Meso Scale Diagnostics, LLC. (MSD), Rockville, MD, USA. **A. Mathew:** A. Employment/Salary (full or part-time); Meso Scale Diagnostics, LLC. (MSD), Rockville, MD, USA. **L. Quintero:** A. Employment/Salary (full or part-time); Meso Scale Diagnostics, LLC. (MSD), Rockville, MD, USA. **J.N. Wohlstadter:** E. Ownership Interest (stock, stock options, royalty, receipt of intellectual property rights/patent holder, excluding diversified mutual funds); Meso Scale Diagnostics, LLC. (MSD), Rockville, MD, USA.

Poster

PSTR180. Biomarkers for Neurodegenerative Diseases

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR180.10/VV60

Topic: I.05. Biomarker, Drug Discovery, and Experimental Therapeutics

Title: The relevance of distinguishing vigilance-sleep states for EEG phenotyping of rodent models and pharmaco-EEG profiling

Authors: *C. HABERMACHER, E. GRONLIER, C. DUMONT, B. CARABALLO, C. ALLIOUX, M. VILLALBA, C. ROUCARD, Y. ROCHE, J. VOLLE;
SynapCell, Saint Ismier, France

Abstract: Sleep and wakefulness are complex and dynamic brain states produced by the interaction of multiple brain structures. Sleep impairments occur also in a wide range of neurodegenerative disorders and could appear during the prodromal phase before the onset of typical clinical symptoms. This suggests the potential of sleep impairments as a biomarker for the early diagnosis of, or as a target of the treatment of these neurodegenerative diseases. Moreover, the identification of the effects of pharmacological compounds on sleep architecture represents a challenge for pharmaceutical industry.Sleep itself is a heterogeneous phenomenon in which two major states can be distinguished: rapid eye movement (REM) sleep and non-rapid eye movement (NREM) sleep. The combination of electroencephalogram (EEG) and electromyogram (EMG) recordings provides objective markers for monitoring wakefulness, NREM sleep and REM sleep. As manual scoring is time-consuming and prone to subjectivity, there is a rising interest in minimizing human intervention. We took advantage of recent advances in artificial intelligence to develop an in-house EEG-based platform to monitor and analyze mouse sleep for drug screening and animal model phenotyping by coupling supervised machine learning and expert scorer analysis.We investigated the relevance of this approach in a context of preclinical drug development. Mice were implanted with parietal and nuchal electrodes, to allow EEG and EMG recordings over 8 hours. Selected reference compounds were tested to assess their effect on sleep architecture and on EEG pattern. Compounds were administered in a cross-over design and recordings were scored in 4 second epochs to identify sleep stages.In this study we identified specific effects of tested compounds. Diazepam, a sedative agent, promoted both sleep states but impacted differently Wake, NREM and REM FFT profiles: it induced a shift towards lower frequencies of REM peak frequency and a reduction of

delta activity (1-4Hz) during NREM stages. Unlike diazepam, the hypnotic agent suvorexant, an orexin receptor antagonist, affected REM more prominently without modifying EEG pattern. Wake-promoting agents like modafinil and caffein suppressed both sleep but impacted EEG power differently.The use of state-dependent frequency bands analysis paves the way to a new dimension in the identification of sleep-wake stages EEG biomarkers. This project lays the foundation for the development of a drug screening process aiming at identifying state-dependent properties of drugs in development or aberrant sleep-vigilance states patterns in pathological models of neurological disorders.

Disclosures: **C. Habermacher:** A. Employment/Salary (full or part-time); SynapCell. **E. Gronlier:** A. Employment/Salary (full or part-time); SynapCell. **C. Dumont:** A. Employment/Salary (full or part-time); SynapCell. **B. Caraballo:** A. Employment/Salary (full or part-time); SynapCell. **C. Allioux:** A. Employment/Salary (full or part-time); SynapCell. **M. Villalba:** A. Employment/Salary (full or part-time); SynapCell. **C. Roucard:** A. Employment/Salary (full or part-time); SynapCell. **Y. Roche:** A. Employment/Salary (full or part-time); SynapCell. **J. Volle:** A. Employment/Salary (full or part-time); SynapCell.

Poster

PSTR180. Biomarkers for Neurodegenerative Diseases

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR180.11/VV61

Topic: I.05. Biomarker, Drug Discovery, and Experimental Therapeutics

Title: Miniscope-based neural circuit profiling in freely behaving animals for preclinical therapeutic assessment

Authors: ***S. HUANG**[1], K. ZITELLI[2], D. CHENG[2], Z. BALEWSKI[2], O. MILLER[2], J. NASSI[2]; [1]Inscopix, Inc., Mountain View, CA; [2]Inscopix, Mountain View, CA

Abstract: Traditional preclinical assays for CNS disorders that rely on behavioral or histological endpoints are often unable to differentiate between distinct mechanisms and have a poor track record of predicting clinical performance. Advancement of effective new therapies into the clinic will be accelerated by more sensitive and predictive assays. The miniscope imaging platform developed by Inscopix allows for cellular resolution activity measurements from hundreds of genetically defined neurons at once. These large-scale neural activity recordings, paired with simultaneous behavioral measurements in freely moving animals, present an opportunity to advance translational research by revealing detailed relationships between neural circuit activity and behavioral symptoms and allowing for the construction of predictive preclinical assays based on the treatment-induced response of large populations of neurons. As a proof of concept, we have established a robust pharmacological dataset with FDA-approved medications for Parkinson's Disease (PD) to facilitate comparison of this approach with traditional behavior-based assays. In 6-OHDA lesioned mice, we have gathered a multidimensional dataset which includes cell-type specific activity of striatal D1 receptor-expressing medium spiny neurons (D1-

MSNs) with synchronized locomotor metrics during free exploration under a breadth of conditions: pre-lesion, post-lesion, therapeutic and dyskinesia-inducing doses of L-DOPA, and dyskinesia-alleviating doses of amantadine. We have observed distinct neural activity patterns across these disease and treatment conditions. We go on to demonstrate that this same approach can be applied to other disease indications, such as Schizophrenia, where we generated a comprehensive striatal D1- and D2-MSN neural circuit profile of a candidate antipsychotic Xanomeline. To further enhance this approach, we present two new applications of the dual-color imaging platform nVue^{TM}, which enables simultaneous imaging of neuronal calcium activity alongside other disease-relevant brain signals such as neurotransmitter (e.g., dopamine) levels or blood flow dynamics. Together, these tools will enable the construction of multidimensional disease and treatment response profiles of neural circuits for basal ganglia-related pathologies such as PD and schizophrenia. Preclinical target engagement and drug efficacy assays informed by these neural profiles promise to be more predictive of clinical performance than current behavior-based assays and should accelerate the development of next-generation therapeutics for a broad array of CNS disorders.

Disclosures: **S. Huang:** A. Employment/Salary (full or part-time); Inscopix. **K. Zitelli:** A. Employment/Salary (full or part-time); Inscopix. **D. Cheng:** A. Employment/Salary (full or part-time); Inscopix. **Z. Balewski:** A. Employment/Salary (full or part-time); Inscopix. **O. Miller:** A. Employment/Salary (full or part-time); Inscopix. **J. Nassi:** A. Employment/Salary (full or part-time); Inscopix.

Poster

PSTR180. Biomarkers for Neurodegenerative Diseases

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR180.12/VV62

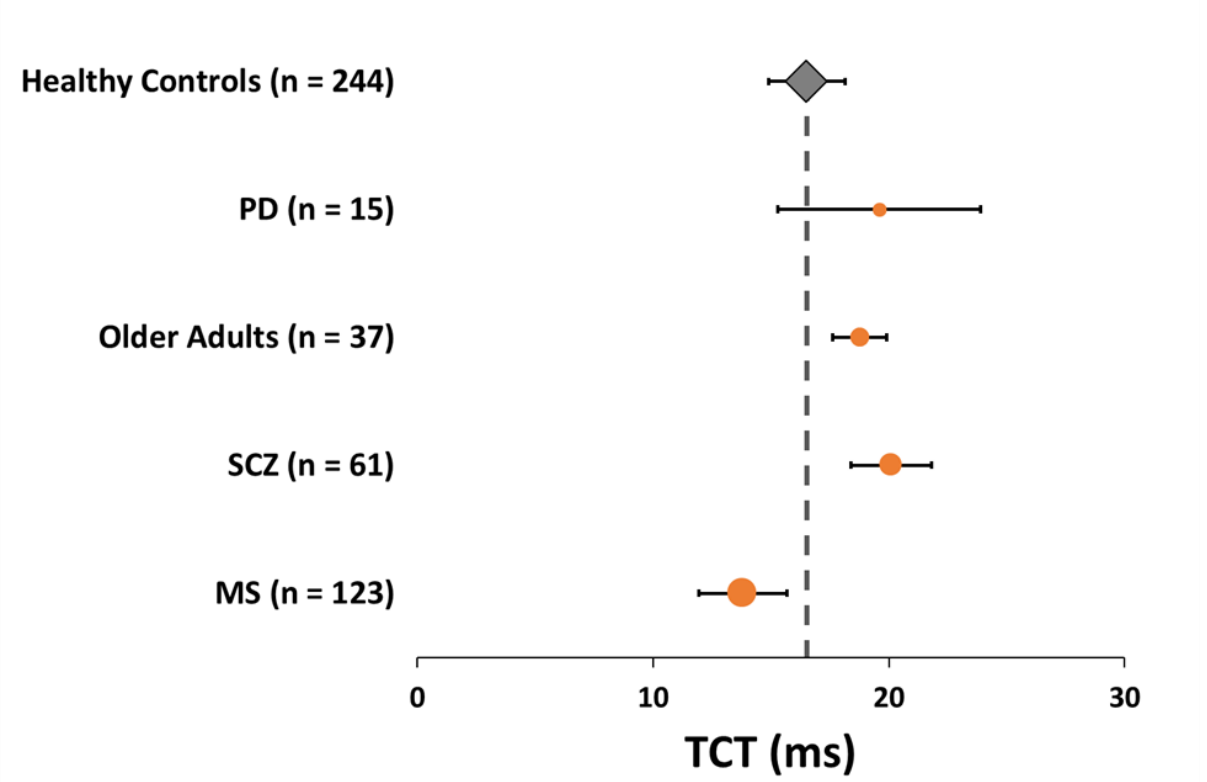
Topic: C.01. Brain Wellness and Aging

Title: Is transcallosal conduction time sensitive to neural disorders? A meta-analysis

Authors: *T. L. DANIELSON, J. M. DEFREITAS;
Kinesiology, Applied Hlth. and Recreation, Oklahoma State Univ., Stillwater, OK

Abstract: Introduction: Transcallosal conduction time (TCT) is a measure of conduction latency across the corpus callosum, specifically in tracts that involve communication between homologous motor cortices. Electromyography is used on a target muscle while transcranial magnetic stimulation (TMS) is applied to both motor cortices independently. The result is an estimate of the time (ms) it takes for a volley to go from one motor cortex to the other. This measure has been proposed to indicate disease or age-related neurodegeneration, though a collection of the normative values found in these populations has not yet been identified. **Methods:** First, a systematic review was conducted that identified any studies that measured TCT. Studies that included a clinical population, as well as healthy controls, were separately identified and used for a meta-analysis of TCT values. The clinical populations with satisfactory

sample sizes or with known associated motor deficits were kept, which included Multiple Sclerosis (MS), Schizophrenia (SCZ), Parkinson's Disease (PD) and aging (OA, for Older Adults). Seventeen studies were included in the meta-analysis with each study's control group contributing to a grand mean of healthy controls, and nine studies contributed TCT values for the clinical populations above. **Results:** The resulting grand means, pooled standard deviations, and total sample sizes for each population are shown in the figure below. **Conclusions:** The SCZ (+3.6 ms), PD (+3.1 ms), and OA (+2.2 ms) populations all showed slower TCT than the control population. The MS population paradoxically has a TCT faster (- 2.7 ms) than the controls. Due to demyelination, we expected MS patients to have slower TCTs. Overall, transcallosal conduction time, as measured via TMS, seems to be sensitive to various neural disorders. However, our review also revealed a concerning diversity of methodologies employed across the studies. For TCT to be useful diagnostically, we believe standardized methods should be developed and widely employed to improve this promising measure.



Disclosures: **T.L. Danielson:** None. **J.M. DeFreitas:** None.

Poster

PSTR181. Neural Stimulation to Understand Brain Function and Dysfunction

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR181.01/VV63

Topic: I.08. Methods to Modulate Neural Activity

Title: Trigeminal Nerve Direct Current Stimulation Modulates Brainstem Nuclei's Activity in Rat

Authors: *A. MAJDI[1], B. ASAMOAH[2], M. MC LAUGHLIN[3];
[1]Deprtment of Neurosci., KU Leuven: Katholieke Univ. Leuven, Leuven, Belgium; [2]Neurosci., [3]KU Leuven, Leuven, Belgium

Abstract: New evidence indicates that the effects of transcranial direct current stimulation (tDCS) on the brain may be indirectly caused by the stimulation of the trigeminal nerve. However, there has been no previous investigation into the electrophysiological effects on the brainstem nuclei when directly stimulating the trigeminal nerve with direct current (DC). These nuclei in turn release neurotransmitters such as serotonin and norepinephrine which can affect function throughout the brain. Male Sprague Dawley rats (n=10) were anesthetized with urethane. DC stimulation was delivered to the lower jaw (0.5 to 3 mA), targeting the marginal branch of the trigeminal nerve. Simultaneously, single-unit electrophysiological recordings (1 minute with no stimulation, 1 minute of DC stimulation, and another 1 minute with no stimulation) were taken from the trigeminal main sensory (NVsnpr), mesencephalic nuclei (MeV), dorsal raphe nucleus (DRN), and median raphe nucleus (MnRN) using a 32-channel silicon probe. A control condition was included where xylocaine was injected to block the trigeminal nerve. A total of 132, 74, 149, and 43 single units were isolated from NVsnpr, MeV, DRN, and MnRN, respectively. DC stimulation of the trigeminal nerve at intensities of 0.5 to 3 mA led to an increase in spiking activity in NVsnpr, MeV, and DRN nuclei. The MnRN single units were categorized into two groups based on their inter-spike interval (ISI) shape: one-peak (type 1, n=18) and multiple-peak (type 2, n=25). Stimulation of the trigeminal nerve with 3 mA caused a decrease in spiking activity in MnRN type 1 cells, and the spike rates remained consistently low after the DC stimulation ended. Conversely, the spike rate of type 2 cells in the MnRN was unaffected by trigeminal nerve DC stimulation. In the xylocaine blocker condition, 20 single units were isolated. DC stimulation of the trigeminal nerve at 3 mA did not induce an increase in spiking activity in the brainstem nuclei. These findings provide the initial evidence that DC stimulation of the trigeminal nerve can modulate the activity in brainstem nuclei. Consequently, these results support the hypothesis that some of the observed effects of tDCS may be attributed to stimulation of cranial nerves.

Disclosures: **A. Majdi:** None. **B. Asamoah:** None. **M. Mc Laughlin:** None.

Poster

PSTR181. Neural Stimulation to Understand Brain Function and Dysfunction

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR181.02/VV64

Topic: I.08. Methods to Modulate Neural Activity

Support: FWO-project grant G0B4520N

Title: Tdcs peripheral nerve stimulation can enhance passive avoidance learning in rats

Authors: *L. VAN BOEKHOLDT[1], S. KERSTENS[1], K. DECLOEDT[2], M. MC LAUGHLIN[3]; [1]Neurosciences, Univ. of Leuven, Leuven, Belgium; [3]Neurosciences, [2]KU Leuven, Leuven, Belgium

Abstract: Transcranial direct current stimulation (tDCS) is a popular non-invasive neuromodulation method that is widely used by neuroscientists and clinicians to modulate brain activity and treat a wide range of disorders. It is assumed that effects are achieved through direct membrane polarization of neurons, but recent research suggests that peripheral nerve activation may be an alternative mode of action. One of the biggest behavioral effects of tDCS in animals is its effect on passive avoidance (PA) learning in rats. Two research groups previously reported that 30 minutes of 0.25mA DC stimulation enhances PA learning. In these experiments, an anodal electrode was placed on the skull (over hippocampus) and a cathodal return electrode on the chest. We set out to investigate whether this increased PA learning was caused by the cerebral or peripheral electric field. Skull electrodes were implanted over the hippocampus and cerebellum while subcutaneous electrodes were implanted over the third occipital nerve and the central back of the animal. Different electrode combinations were used to separate the cerebral and peripheral electric fields in four different experimental groups (n= 16 per group): skull-only, subcutaneous-only, skull-subcutaneous, and sham. After 30 minutes of 0.25 mA (or sham) stimulation, the training session of the PA task was performed. One day later, PA learning was assessed in a testing session. Analysis revealed that PA learning, as assessed by step-through latency, was not increased in any of the stimulation groups compared to sham. The used amplitude of 0.25 mA is lower than amplitudes generally used in human tDCS to correct the resulting cerebral electric field for the thinner rat skull. This will however also lower the electric field strength applied to the skin. In a follow-up experiment, we therefore investigated whether 30 minutes of subcutaneous stimulation at 2mA effected PA learning. Analysis revealed a significant increase in the step-through latency of the subcutaneous stimulation group compared to the sham group ($p = 0.048$; $n = 13$ per group). This finding is particularly relevant to human tDCS as the used amplitude of 2mA is also one of the standards applied in human tDCS stimulation paradigms, providing additional evidence that peripheral nerve activation could be an alternative mode of action. The cathodal electrode in our last experiment was positioned over the third occipital nerve. This nerve that is also stimulated in many conventional human tDCS montages, suggesting that occipital nerve activation could be at the basis of some effects. Future experiments are necessary to investigate occipital nerve involvement in human tDCS effects.

Disclosures: L. van Boekholdt: None. **S. Kerstens:** None. **K. Decloedt:** None. **M. Mc Laughlin:** None.

Poster

PSTR181. Neural Stimulation to Understand Brain Function and Dysfunction

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR181.03/WW1

Topic: I.08. Methods to Modulate Neural Activity

Support: NIH Grant 1R01MH123508-01

Title: The role of the LC-NE system in external trigeminal nerve stimulation, a pupil study

Authors: *N. SEMINCK[1], A. KHATOUN[1], B. NUTTIN[2], M. MC LAUGHLIN[1];
[1]Neurosciences, [2]Neurosurg., KU Leuven, Leuven, Belgium

Abstract: Background: External trigeminal nerve stimulation (eTNS) is a non-invasive neuromodulation technique that is being explored for the treatment of epilepsy, major depression disorder, and stress-related disorders. It has already been approved as a treatment for migraines and ADHD in children. Despite the clinical efficacy of eTNS, the underlying mechanisms remain poorly understood. The trigeminal nerve provides input to various brainstem nuclei, which subsequently activate the locus coeruleus (LC), regulating the levels of norepinephrine (NE). LC activity has been linked to pupil dilation, while NE is involved in processes such as arousal and memory retrieval. Therefore, the LC-NE system may play a crucial role in the mechanism of eTNS.
Objective: We aimed to investigate the effect of eTNS on acute pupil responses in healthy volunteers. We hypothesized that eTNS would induce pupil dilation through activation of the LC-NE system.
Methods: Twenty participants underwent three stimulation conditions (eTNS, sham, and median nerve stimulation - MNS), while pupil diameter was measured. MNS was included as an additional control as it is believed to not activate the LC-NE pathway. eTNS, sham, and MNS were applied alternatingly with a 4000 ms interval between each stimulation condition.
Results: Both eTNS and MNS resulted in larger pupil responses (approximately 1.5ms after stimulation onset) compared to sham. However, eTNS produced the largest pupil dilation, and the number of pupil dilations was significantly higher after eTNS compared to MNS and sham. The latency of the pupil response did not differ between eTNS and MNS, but was significantly delayed in the sham condition. The size of the pupil response was correlated with the baseline pupil diameter.
Conclusion: eTNS induces significantly greater pupil dilation compared to the MNS-control stimulation. These findings suggest that eTNS modulates the release of NE, leading to pupil dilation. Consequently, the LC-NE system may serve as a crucial mediator of the effects of eTNS.

Disclosures: **N. Seminck:** None. **A. Khatoun:** None. **B. Nuttin:** None. **M. Mc Laughlin:** None.

Poster

PSTR181. Neural Stimulation to Understand Brain Function and Dysfunction

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR181.04/WW2

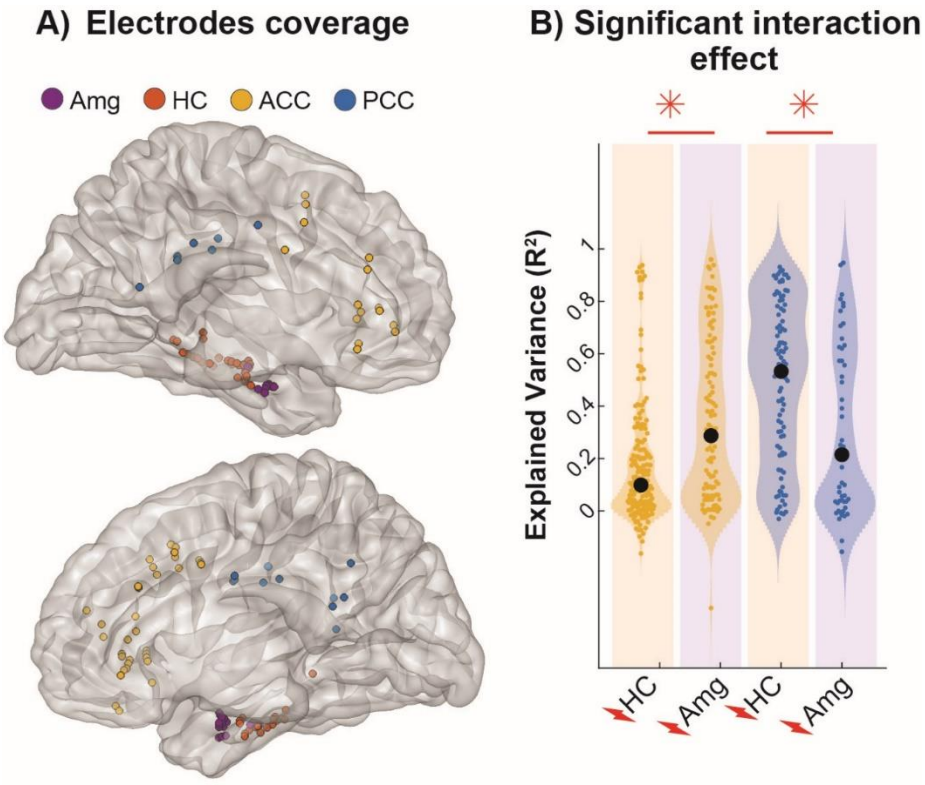
Topic: I.08. Methods to Modulate Neural Activity

Support: RO1 MH122258

Title: Limbic subsystems are strongly intra-connected

Authors: *G. OJEDA VALENCIA[1], N. M. GREGG[2], H. HUANG[3], B. N. LUNDSTROM[2], B. H. BRINKMANN[2], T. PAL ATTIA[4], J. VAN GOMPEL[5], M. A. BERNSTEIN[6], M.-H. IN[6], J. I. HUSTON[7], G. A. WORRELL[8], K. J. MILLER[5], D. HERMES[9];
[1]Physiol. and BiomedicalEngineering, [2]Neurol., [3]Mayo Clin. Med. Scientist Training Program, [5]Neurosurg., [6]Radiology, [7]Rediology, [8]of Physiol. and Biomed. Engineering, Neurol., [9]Physiol. and Biomed. Engineering, Radiology, Neurol., [4]Mayo Clin., Rochester, MN

Abstract: Emotion and memory in the human brain are supported by the limbic network. Previous anatomical and functional studies have shown at least two subnetworks within the limbic system, which is often targeted for brain stimulation. Limbic targets for stimulation therapies are investigated for the treatment of depression and Alzheimer's disease. As stimulation affects the network, it is essential to understand to what degree subsystems can be selectively modulated. In this study, we therefore used single pulse stimulation and measured cortico-cortical evoked potentials (CCEPs) to test whether intra-network connections within limbic subsystems were stronger compared to inter-network connections. In 8 patients with drug resistant epilepsy we stimulated the limbic system to measure CCEPs between the hippocampal complex (HC), amygdala (Amg), posterior cingulate cortex (PCC), anterior cingulate cortex (ACC). Using a novel Canonical Response Parameterization (CRP) method we characterized the reliability and amplitude of CCEPs. This method allows us to investigate long latency CCEPs (500 ms), likely related to indirect responses, in addition to short latency responses (<50 ms) that have been related to direct projections. We used a linear mixed effects model to test whether connections within subsystems (Amg-ACC, HC-PCC) were more reliable compared to connections between subsystems (HC-ACC, Amg-PCC).The linear mixed effects model showed that Amg and HC stimulation had different effects on ACC and PCC (significant interaction, $t(440) = 6.109, p=2.2*10^{-9}$). Further analysis showed that CCEPs in the ACC were significantly more reliable with Amg compared to HC stimulation ($t(294) = -3.829, p=3.2*10^{-4}$, *2 post-hoc tests, Bonferroni corrected*). Conversely, CCEPs in the PCC were significantly more reliable with HC compared to Amg stimulation ($t(146) = 5.011, p=3.1*10^{-6}$, *2 post-hoc tests, Bonferroni corrected*). Using CRP, we studied the network wide effects of stimulation in the limbic system. Limbic subsystems have more reliable intra-network compared to inter-networks connectivity



Disclosures: G. Ojeda Valencia: None. **N.M. gregg:** B. Contracted Research/Research Grant (principal investigator for a drug study, collaborator or consultant and pending and current grants). If you are a PI for a drug study, report that research relationship even if those funds come to an institution.; Coinvestigator for the Medtronic EPAS trial, Industry consultant for NeuroOne, funds to Mayo Clinic. **H. Huang:** None. **B.N. Lundstrom:** B. Contracted Research/Research Grant (principal investigator for a drug study, collaborator or consultant and pending and current grants). If you are a PI for a drug study, report that research relationship even if those funds come to an institution.; Site investigator for Medtronic EPAS, Site investigator for NeuroPace RESPONSE, Site investigator for Neuroelectrics tDCS for Epilepsy, Industry consultant for Epiminder, funds to Mayo Clinic, Industry consultant for Medtronic, funds to Mayo Clinic, Industry consultant for Neuropace, funds to Mayo Clinic, Industry consultant for Philips Neuro, funds to Mayo Clinic. E. Ownership Interest (stock, stock options, royalty, receipt of intellectual property rights/patent holder, excluding diversified mutual funds); Intellectual property licensed to Cadence Neuroscience Inc (contractual rights waived), Intellectual property licensed to Seer Medical Inc (contractual rights waived). **B.H. Brinkmann:** E. Ownership Interest (stock, stock options, royalty, receipt of intellectual property rights/patent holder, excluding diversified mutual funds); Intellectual property licensed to Cadence Neuroscience Inc (contractual rights waived), Site investigator for Medtronic EPAS trial. **T. Pal Attia:** None. **J. Van Gompel:** B. Contracted Research/Research Grant (principal investigator for a drug study, collaborator or consultant and pending and current grants). If you are a PI for a drug study, report that research relationship even if those funds come to an institution.; Coinvestigator for SLATE trial, Coinvestigator for the Medtronic EPAS trial, Coinvestigator for Mayo Clinic Medtronic NIH Public Private Partnership (UH3-NS95495), site Primary

Investigator in the Polyganics ENCASE II trial,, site Primary Investigator in the NXDC Gleolan Men301 trial,, site Primary Investigator in the Insightec MRgUS EP001 trail;. E. Ownership Interest (stock, stock options, royalty, receipt of intellectual property rights/patent holder, excluding diversified mutual funds); Intellectual property licensed to Cadence Neuroscience Inc, co-owned by Mayo Clinic, Stock Ownership and Consulting Contract with Neuro-One Inc,. **M.A. Bernstein:** A. Employment/Salary (full or part-time); former employee of GE Medical Systems and receives pension payments.. **M. In:** None. **J.I. Huston:** None. **G.A. Worrell:** E. Ownership Interest (stock, stock options, royalty, receipt of intellectual property rights/patent holder, excluding diversified mutual funds); declares intellectual property disclosures related to behavioral state, seizure detection and seizure forecasting algorithms, Intellectual property licensed to Cadence Neuroscience Inc. and NeuroOne Inc. **K.J. Miller:** None. **D. Hermes:** None.

Poster

PSTR181. Neural Stimulation to Understand Brain Function and Dysfunction

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR181.05/WW3

Topic: I.08. Methods to Modulate Neural Activity

Support: NIH P41-EB018783
NIH/NIMH R01-MH120194
NIH/NIBIB R01-EB026439
NIH/NINDS U24-NS109103
NIH/NINDS U01-NS108916
NIH/NINDS U01-NS128612
McDonnell Center for Systems Neuroscience
Fondazione Neurone
AES Grant 980019

Title: Mapping cortical responses to thalamic stimulation in human during stereo-electroencephalography evaluation

Authors: *X. LIU[1,2], L. N. EISENMAN[3], P. BRUNNER[2,3,4,5], J. T. WILLIE[2,3,4,5];
[1]Washington Univ. in St. Louis, Saint Louis, MO; [2]Dept. of Neurosurg., [3]Dept. of Neurol., Washington Univ. Sch. of Med. in St Louis, St. Louis, MO; [4]Dept. of Biomed. Engin., Washington Univ. in St Louis, St. Louis, MO; [5]Natl. Ctr. for Adaptive Neurotechnology, St. Louis, MO

Abstract: Deep brain stimulation of the thalamus is an emerging option to treat drug-resistant epilepsy. Stimulation candidates include anterior (ANT), centromedian (CM), and pulvinar medial (PuM) nuclei of the thalamus, which can disrupt epileptic activity via perturbation of thalamocortical loops. We set out to map cortical and subcortical responses to single pulse electrical stimulation (SPES) of ANT, CM, and PuM in three patients with presumed temporal

lobe epilepsy who underwent invasive stereo-electroencephalography (sEEG) monitoring. The post-operative electrode localization, cortical parcellation, and thalamic segmentations were performed with FreeSurfer. We analyzed two leads implanted in left ANT, one lead in left CM and left PuM respectively across three subjects. To probe functional connectivity between the thalamus and cortex, SPES was delivered through thalamic contacts (60 bi-phasic pulses, 200us pulse duration, 3 and 6 mA intensities, 60 trials at 0.5Hz per intensity). Evoked potentials were computed by averaging signals time-locked to stimulation onset. To identify contacts responding to thalamic stimulation, we used Wilcoxon rank sum tests to determine if the distribution of inter-trail correlations of signal 300 ms before and after stimulation are significantly different ($p < 0.05$, responder contact). We found dose-dependent responses to thalamic stimulation at 6 mA to yield stronger responses. Contacts that responded to stimulation of PuM were mainly located within the inferior temporal lobe. CM stimulation revealed cortical and subcortical projections, including the cingulate cortex, putamen, pallidum, frontal and parietal lobes, precentral, postcentral, and paracentral gyrus. ANT stimulation evoked prominent responses in the inferior and middle temporal regions in two patients. This study provides a systematic framework to study effective functional connectivity between thalamic neuromodulation targets and the cortex. The insight gained from this study can help in the investigation of the role of thalamocortical connections in the propagation of epileptic activity.

Disclosures: X. Liu: None. **L.N. Eisenman:** None. **P. Brunner:** None. **J.T. Willie:** None.

Poster

PSTR181. Neural Stimulation to Understand Brain Function and Dysfunction

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR181.06/WW4

Topic: I.08. Methods to Modulate Neural Activity

Support: SNF Grant 197766
R01-2NS062092
CDMRP W81XWH-22-1-0315

Title: Anterior and Centromedian Thalamic Nuclei: Identifying Thalamic Connectivity for Optimizing Target Selection in Neuromodulation for Epilepsy

Authors: *G. AIELLO[1,2], L. IMBACH[3], R. POLANIA[4], S. S. CASH[2], P. SALAMI[2];
[1]Swiss Federal Inst. of Technol. Zurich, Zurich, Switzerland; [2]Massachusetts Gen. Hosp., Boston, MA; [3]Swiss Epilepsy Clin., Zurich, Switzerland; [4]ETH Zurich, Zurich, Switzerland

Abstract: The Anterior (ANT) and Centromedian (CM) thalamic nuclei are key targets for neuromodulatory treatments in epilepsy, with the ANT being mostly studied for focal epilepsy, while the CM for generalized epilepsy. However, the optimal target selection and varied patient responses remain unclear. Recent research highlights the significance of θ and low β oscillations in determining responsiveness to ANT deep brain stimulation (DBS), therefore this study

investigates the dynamic functional interaction of ANT and CM with cortical regions in these frequency bands to differentiate their roles in different epilepsy types. Multi-day interictal recordings during the presurgical evaluation of nine patients were analyzed. The spectral properties of ANT (N=6) and CM (N=6) as well as their interaction with the scalp electrodes were examined using weighted phase lag index debiased. Statistical analyses, including ANOVA with multiple comparisons correction, were conducted. We found that CM power exceeded ANT in lower frequencies (δ and θ, $p<0.001$), but the opposite was true for higher frequencies. Subject variability was most pronounced in θ power, consistent with our previous findings, suggesting its importance in predicting response to thalamic neurostimulation. Furthermore, the greatest difference between CM and ANT connectivity to the cortex was concentrated in θ and low β frequencies, with CM showing dominance in θ, while ANT connectivity to the scalp being more prominent in low β. CM exhibited stronger connectivity than ANT in a widespread range of cortical regions, exclusively in the θ band, suggesting its suitability for generalized epilepsies. Conversely, ANT demonstrated stronger connectivity to the cortex in the low β band, particularly in the posterior-temporal region, indicating its relevance in treating focal epilepsies, including temporal lobe involvement. Recent findings support this, showing stronger low β ANT-scalp connectivity in responders to ANT DBS compared to non-responders. These findings highlight significant spectral and connectivity differences between ANT and CM, the two primary neuromodulation targets for epilepsy. We suggest that frequency-specific differential connectivity profiles may contribute to varied responses in ANT and CM stimulation. We believe that ANT could target low β oscillations, impacting focal, temporo-posterior cortex, while CM influences widespread cortical activity via θ oscillations, making it suitable for generalized epilepsies. These mechanisms may guide future studies in identifying optimal thalamic targets and stimulation parameters for epilepsy.

Disclosures: G. Aiello: None. **L. Imbach:** None. **R. Polania:** None. **S.S. Cash:** None. **P. Salami:** None.

Poster

PSTR181. Neural Stimulation to Understand Brain Function and Dysfunction

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR181.07/WW5

Topic: I.08. Methods to Modulate Neural Activity

Title: Influence of intracortical microstimulation on synaptic efficacy between visual and executive cortical areas in macaques

Authors: *S. D. EGRANOV[1], R. MILTON[2], M. SLAPIK[3], V. DRAGOI[4];
[1]Neurobio. & Anat., UTHealth Houston Neurosci. Grad. Program, Houston, TX; [2]Neurobio. and Anat., Univ. of Texas Hlth. Sci. Ctr. At Houst, Houston, TX; [3]McGovern Med. Sch., Houston, TX; [4]Dept Neurobiol/Anat, Univ. Texas-Houston Med. Sch., HOUSTON, TX

Abstract: Intracortical microstimulation (ICMS) technologies have been previously utilized as a means to elucidate the circuitry underlying distinct regions of the cortex, as well as the functions of their respective neural populations. Despite the emergence ICMS tools for externally driving neuron spiking activity, the nature of the cortico-cortical connectivity between neuron populations in the visual cortex and the prefrontal cortex and the pathways for communication between these brain areas demand further study. To uncover alterations in synaptic efficacy and neural firing biases between visual and decision-making cortical regions, we performed high-yield electrophysiological recordings simultaneously from area V4, a mid-tier visual cortical region, and the dorsolateral prefrontal cortex (dlPFC). We investigated how neural activity within and between each area was affected by the induction of an externally-derived electrical current and directly probed the communication pathways between mid-tier visual processing and decision-making areas. ICMS of neuron subpopulations within each respective region produced distinct alterations in pre-synaptic and post-synaptic spiking activity both within and between our cortical areas of interest, suggesting modulation of synaptic efficacy in dlPFC and V4 following microstimulation. Our study provided key insights into the circuitry linking decision-making and visual areas and ascertained the functional connectivity and neural coupling of both local and distal neurons within and between visual area V4 and dlPFC.

Disclosures: **S.D. Egranov:** None. **R. Milton:** None. **M. Slapik:** None. **V. Dragoi:** None.

Poster

PSTR181. Neural Stimulation to Understand Brain Function and Dysfunction

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR181.08/WW6

Topic: I.08. Methods to Modulate Neural Activity

Support: NIH Grant R01NS109361
NIH Grant R01NS102917
NIH Grant U01 NS115588

Title: Neuronal dynamics underlying behavioral detection in chronic intracortical microstimulation

Authors: *R. KIM[1], R. LYCKE[1], J. MONTES[2], C. XIE[1,2], L. LUAN[1,2];
[1]Electrical and Computer Engin., [2]Bioengineering, Rice Neuroengineering Initiative, Houston, TX

Abstract: Intracortical microstimulation (ICMS) uses microelectrodes to activate neurons through the delivery of electrical current. It holds promise for prosthetic applications that can restore lost functions such as sensation of touch or sight. However, the precise neuronal mechanism underlying the behavioral detection of ICMS and the effects of various factors such as implantation duration and stimulation paradigms are poorly understood. Several challenges, including the difficulty to simultaneously stimulate, record large-scale neuronal activation, and

perform behavior tasks, coupled with the instability of the electrode-tissue interface, have hindered the understanding of neuronal dynamics underlying behavioral detection in ICMS. We present the application of StimNET, an ultraflexible stimulation electrode, alongside a comprehensive set of imaging and behavioral methods to overcome these challenges. To investigate the relationship between neuronal activation and behavioral responses, we trained water-deprived, head-fixed mice in a go/no-go task to turn a wheel beyond an angular threshold in response to ICMS to obtain water reward. Simultaneous stimulation and volumetric Ca^{2+} imaging of neuronal activation during longitudinal behavioral testing were achieved by co-implanting a cranial window with StimNET in Thy1-GCamp6s mice.Our findings reveal intriguing relationships between neuronal activation and behavioral detection threshold. Over chronic periods, the number of ICMS activated neurons tracked the change of behavioral detection threshold, with greater population of activation at the initial phase compared to that of the later stable phase. At identical threshold currents that elicited both go and no-go responses, the Ca^{2+} fluorescence associated with neuronal activation peaked faster for go responses compared to no-go responses, while the number of activated neurons remained similar. Furthermore, altering the stimulation pulse width and frequency modulated the underlying neuronal activation at the behavioral detection threshold. These results provide valuable insights into the driving factors of the diverse neuronal responses underlying the same behavioral detection, offering guidance for optimizing microstimulation paradigms to enhance behavior detectability.

Disclosures: **R. Kim:** None. **R. Lycke:** None. **J. Montes:** None. **C. Xie:** E. Ownership Interest (stock, stock options, royalty, receipt of intellectual property rights/patent holder, excluding diversified mutual funds); Neuralthread Inc. **L. Luan:** E. Ownership Interest (stock, stock options, royalty, receipt of intellectual property rights/patent holder, excluding diversified mutual funds); Neuralthread Inc.

Poster

PSTR181. Neural Stimulation to Understand Brain Function and Dysfunction

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR181.09/WW7

Topic: I.08. Methods to Modulate Neural Activity

Support: NIH/NINDS (R01-NS090874)
NIH/NINDS (R01-NS101013)
NIH/NIBIB (R01-EB026439)
NIH/NINDS (U24-NS109103)
NIH/NINDS (U01-NS108916)
NIH/NIMH (R01-MH120194)
NIH/NINDS (F32-NS124837)
NIH/NINDS (R25-NS090978)
NREF FAANS, and the AANS/CNS Cerebrovascular Section
McDonnell Center for Systems Neuroscience

Title: Effect of anesthesia-induced burst suppression on cortico-cortico evoked potentials

Authors: *A. SIMMONS[1,2], M. ADAMEK[2,3], A. I. SRIENC[2], A. AGATO[1,4], S. A. ANAND[1], P. DEMAREST[1,2], T. XIE[2,5], D. W. MORAN[1], J. P. CULVER[1,4], J. T. WILLIE[1,2,5], P. BRUNNER[1,2,5];
[1]Biomed. Engin., [2]Neurosurg., [3]Neurosci., [4]Radiology, Washington Univ. in St. Louis, St. Louis, MO; [5]Natl. Ctr. for Adaptive Neurotechnologies, St. Louis, MO

Abstract: Anesthesia-induced loss of consciousness is known to affect neuron excitability and, consequently, the brain's state of activity. Under deep anesthesia, a pattern of semi-periodic neuronal bursting followed by periods of suppressed activity called burst suppression is widely observed across subjects. While anesthesia depth has been observed to correlate with changes in evoked potential (EP) characteristics, the separate impact of the bursting and suppression states observed during anesthesia-induced burst suppression on EPs is not well characterized. The aim of this study is to determine the effect of anesthesia-induced burst suppression on cortico-cortical evoked potentials (CCEPs). Specifically, this work seeks to investigate whether CCEPs, measured during bursting (up states), differ characteristically from CCEPs measured during suppression (down states). We hypothesize that during up states, CCEPs exhibit higher peak-to-peak amplitudes than during down states. To investigate this hypothesis, we analyzed CCEP data recorded in two non-human primates (NHP). The NHPs were implanted with 12 SEEG depth electrodes across the visual cortex. Throughout the experiment, the NHPs were kept anesthetized, and burst suppression was visible in the SEEG recordings. Single pulse electrical stimulation was delivered at a rate of 0.5 Hz for approximately 2 minutes (60 trials) between two contacts in visual area 1. The other contacts were used to record the evoked response across the visual cortex. For this analysis, the signals were first detrended to minimize electrode drift. Next, each stimulation trial was visually classified as having occurred during an up or down state. Half of the trials occurred during up states and half during down states. We compared, for each contact, the average peak-to-peak amplitude and latency of the responses during up and down states. Out of 48 total contacts, 43 recorded larger responses during up states. We found no relationship between response latency and up/down state classification. This result aligns with our hypothesis that CCEP amplitude may be affected by burst suppression. It is possible that the semi-periodic up states that appear during burst suppression are indicative of simultaneous, semi-periodic increases in brain connectivity and/or excitability. Consequently, the results of this investigation may have significance in improving our mechanistic understanding of how and why burst suppression arises.

Disclosures: A. Simmons: None. **M. Adamek:** None. **A.I. Srienc:** None. **A. Agato:** None. **S.A. Anand:** None. **P. Demarest:** None. **T. Xie:** None. **D.W. Moran:** None. **J.P. Culver:** None. **J.T. Willie:** None. **P. Brunner:** None.

Poster

PSTR181. Neural Stimulation to Understand Brain Function and Dysfunction

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR181.10/WW8

Topic: I.08. Methods to Modulate Neural Activity

Title: Temporal interference brain stimulation to the dorsomedial prefrontal cortex alters Stroop effect

Authors: *J. RYAN[1], B. BOTZANOWSKI[1], M. KARKARE[1], T. FULTON[1], J. KUBERT[1], S. LIU[1], S. BETTERS[1], A. WILLIAMSON[2], N. FANI[1], M. TREADWAY[1];
[1]Emory Univ., Atlanta, GA; [2]Aix-Marseille Univ., Marseille, France

Abstract: The identification and understanding of the mechanistic underpinnings of human behavior are crucial for the advancement of neuroscience and the development of new clinical treatments. Utilization of brain stimulation techniques, such as transcranial magnetic simulation and transcranial alternating current stimulation, offer a means to manipulate brain regions in a non-invasive manner, but suffer from focality and depth limitations. Attempts to resolve these issues led to the development of Temporal Interference (TI), a non-invasive brain stimulation that utilizes temporally interfering electric fields to directly stimulate regions at any depth and with great precision, offering the opportunity to garner novel insights of targeted structures and related mechanisms. For this study, healthy participants were recruited and administered TI-on (20 Hz envelope) and TI-off stimulations (0 Hz envelope) in interleaved blocks while completing two versions of the Stroop Task, a well-established paradigm of cognitive control. Stimulations targeted the dorsomedial prefrontal cortex (dmPFC), a brain region associated with cognitive control and related to cognitive dysfunction in multiple psychiatric disorders. For the color-word Stroop Task, participants were instructed to press a button corresponding to the color of the word while ignoring the semantic meaning. Similarly, for the affective Stroop Task, participants were instructed to press a button corresponding to the quantity of numbers while ignoring their numerical values. For the affective Stroop, participants were also presented with positive, aversive, or neutral distractor stimuli before and after presentation of a number stimulus array to assess whether distractor valence affects response latency. For both tasks, mean response time differences between congruent (e.g., “red” written in red or five instances of “5”) and incongruent trials (e.g., “red” written in green or five instances of “3”) within each block were used to assess cognitive control (i.e., the Stroop Effect). Results revealed 20 Hz dmPFC TI led to a reduced Stroop effect compared to the dmPFC TI-off stimulation when controlling for order effects. Additionally, this effect was largely driven by a reduced response time to incongruent trials, suggesting enhanced performance due to changes in dmPFC cognitive processing. These findings are some of the first to measure behavioral changes following TI within human subjects and supports claims of TI as an effective tool to investigate brain regions and related mechanisms, as well as its clinical potential through modulation of areas associated with brain dysfunction.

Disclosures: **J. Ryan:** None. **B. Botzanowski:** None. **M. Karkare:** None. **T. Fulton:** None. **J. Kubert:** None. **S. Liu:** None. **S. Betters:** None. **A. Williamson:** None. **N. Fani:** None. **M. Treadway:** None.

Poster

PSTR181. Neural Stimulation to Understand Brain Function and Dysfunction

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR181.11/WW9

Topic: I.08. Methods to Modulate Neural Activity

Support: European Union's Horizon 2020 research and innovation programme 2021-2024 under grant agreement number 101017716 ("Neurotwin")

Title: Induction and stabilization of gamma oscillations in the human brain

Authors: *B. GLINSKI[1,2], M. SALEHINEJAD[1], K. TAKAHASHI[1,2], M.-F. KUO[1], M. A. NITSCHE[1,3];
[1]Leibniz Res. Ctr. for Working Envrn. and Human Factors (IfADo), Dortmund, Germany; [2]Dept. of Psychology, Ruhr-University Bochum, Bochum, Germany; [3]Dept. of Neurol., Univ. Med. Hosp. Bergmannsheil, Bochum, Germany

Abstract: Alzheimer's disease (AD) constitutes a serious burden on the global health system. According to the World Health Organization (WHO) dementia cases, including AD, will triple by 2050. However, treatment approaches are limited. Accumulated evidence suggests non-pharmacological interventions such as non-invasive brain stimulation (NIBS) technologies e.g., transcranial alternating current stimulation (tACS) or repetitive transcranial magnetic stimulation (rTMS), especially in the gamma frequency range, as a potential treatment approach for underlying causes of AD. The efficacy of these techniques is unclear and requires systematic exploration. The present randomized, crossover, single-blinded study aimed to develop novel NIBS protocols for the induction and stabilization of gamma oscillations in the human cortex of 30 healthy participants. In total five NIBS protocols were explored. These protocols included (1) a gamma frequency intermittent theta burst protocol (gTBS), (2) a gamma frequency tACS protocol, (3,4) two gTBS protocols phase-locked to the peak or the trough of the tACS-generated sinusoidal wave (Peak/Trough X-tACS) and (5) a Sham protocol. The effects of the protocols were evaluated via resting-state electroencephalography and behavioral working memory performance (N-Back task). The results show an increase in gamma power (40Hz) for the tACS protocol lasting up to two hours after stimulation compared to the sham condition ($p<0.001$). Additionally, a short-lasting increase of 40Hz oscillations was found for the gTBS protocol lasting for up to 30 minutes after stimulation compared to the sham condition ($p<0.05$). A comparison to the respective baseline showed a short-lasting (up to 30 minutes) increase of 40Hz power for the X-tACS Peak, gTBS, and tACS conditions ($p´s<0.005$), and a significant increase in all conditions compared to the respective baselines ($p<0.05$) from 60-120 minutes after stimulation. This latter effect might be attributable to changes in brain states due to the two hours of after-measurements. During working memory task performance, a significantly decreased reaction time was observed only for the tACS and gTBS conditions in the 3-Back condition compared to the sham condition and to the respective baseline performance ($p <0.05$). No effects on task performance accuracy were observed. No effects were found for the 1-Back working memory task condition. The results suggest the effectiveness of different non-invasive brain stimulation for the entrainment of gamma oscillations in the healthy human cortex. These effects nominate NIBS as a promising approach in pathological populations.

Disclosures: **B. Glinski:** None. **M. Salehinejad:** None. **K. Takahashi:** None. **M. Kuo:** None. **M.A. Nitsche:** F. Consulting Fees (e.g., advisory boards); Member of the advisory board of Neuroelectrics.

Poster

PSTR181. Neural Stimulation to Understand Brain Function and Dysfunction

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR181.12/WW10

Topic: I.08. Methods to Modulate Neural Activity

Support: ERC

Title: Competition between auditory rhythms and electrical stimulation to modulate behavior

Authors: *Y. CABRAL-CALDERIN[1], M. J. HENRY[2,3];
[1]MPI for Empirical Aesthetics, Frankfurt am Main, Germany; [2]Max Planck Inst. for Empirical Aesthetics, Frankfurt Am Main, Germany; [3]Toronto Metropolitan Univ., Toronto, ON, Canada

Abstract: Neural tracking (entrainment) of auditory rhythms benefits perception. One promising approach to improve this tracking is brain stimulation. We previously demonstrated that frequency-modulated (FM) sounds entrain neural activity and influence the perception of faint auditory targets. Additionally, when combined with transcranial alternating current stimulation (tACS), entrainment to the FM sound can be either enhanced or suppressed, depending on the timing between the electrical and auditory signals. However, tACS effects appear to be primarily modulatory as the FM stimulus still exerts a stronger influence on behavior. Here, we delve deeper into this phenomenon by investigating the interaction between entrainment to tACS, and auditory rhythms. We hypothesized that the effects of tACS would be more pronounced when auditory stimuli were less strongly modulated, as the competition between the two signals would be weaker. In two separate experiments, participants were presented with FM stimuli with varying levels of modulation depth: 39% and 11% (Experiment 1, N=34) and 0% (Experiment 2, N=25). Their task was to detect silent gaps embedded within sounds. During the task, tACS was applied targeting auditory regions either at the same FM stimulus rate (2 Hz in Experiment 1) or at different frequencies (0.8, 2, 3.2, and 4.4 Hz Experiment 2). Results in Experiment 1 showed that behavioral entrainment to sounds was weaker for stimuli with 11% modulation depth compared to 39%. As expected, tACS influenced the strength of behavioral entrainment to the auditory stimulus. However, contrary to our hypothesis, the modulation depth of the auditory stimulus did not affect the magnitude of tACS effects. In Experiment 2, where no rhythmic information was conveyed by the sound, tACS significantly modulated the detection of the near-threshold auditory target in a sinusoidal manner. While group-level analysis did not reveal a significant effect of tACS frequency, clustering analysis identified three distinct groups of participants based on the strength of the tACS effect for different stimulation frequencies. Interestingly, an oscillator model with a free parameter for the individual resonance frequency produced similar response profiles. Our findings suggest that when both sensory and electrical

stimuli are rhythmic, sensory stimuli prevail in entraining behavior and tACS effects become purely modulatory. When no competition occurs between sensory and electrical stimulations, the effects of tACS depend on the individual's preferred frequency, highlighting the importance of accurately targeting individual frequencies during tACS experiments.

Disclosures: Y. Cabral-Calderin: None. **M.J. Henry:** None.

Poster

PSTR181. Neural Stimulation to Understand Brain Function and Dysfunction

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR181.13/WW11

Topic: I.08. Methods to Modulate Neural Activity

Support: 1R01NS113782-01A1

Title: Functional MRI monitoring of non-invasive deep brain stimulation in rats reveals anatomical shunting

Authors: *M. VOROSLAKOS[1,2], T. M. AREFIN[3], J. ZHANG[3], L. ALON[3], D. K. SODICKSON[3], G. BUZSAKI[2];
[1]NYU, New York, NY; [2]Neurosci. Inst., [3]Dept. of Radiology, NYU Grosmann Sch. of Med., New York, NY

Abstract: Transcranial Electrical Stimulation (TES) is a noninvasive method that can modulate neuronal activity. Its ability to limit the effects of stimulation to a target region while minimizing the currents in non-target areas is challenging (i.e., focusability), because the electric fields expected to be highest in superficial cortical regions underneath the stimulating electrodes and decreasing progressively as a function of distance from the cathode. However, experimental evidence for this assumption is lacking. Brain areas distal from the stimulation electrodes might be affected by the inhomogeneous conductivity of the brain, particularly current shunts via ventricles. To measure the whole brain effects of TES, we combined neurostimulation with electrophysiology and BOLD fMRI in urethane anesthetized rats. We measured the TES-induced neuronal activity in the auditory cortex (i.e., below the cathode and anode), CA1 and CA3 regions of the hippocampus and thalamus in the same rat by repeated penetrations with a 4-shank Neuropixel 2.0 probe (total of 1536 sites). We acquired fMRI data with T2*-weighted single-shot GE-EPI sequence. The rat brain, excluding the cerebellum, was covered using 23 axial slices with the following parameters: TE/TR=13.4/(1500 or 2200)ms (TES or resting state fMRI), 300 repetitions and resolution = 0.23 x 0.23 x 0.8mm. 500-ms TES pulses were applied at various intensities (50, 100 and 250 μA), and frequencies (direct current pulses and 2, 4 and 8 Hz alternating current pulses). To validate our MRI data, we performed electrophysiology recording of spiking responses from the hippocampus after the fMRI experiments.In our electrophysiology experiments, we found that electric fields did not decrease monotonically with distance from the stimulation electrodes but showed decreased field around the lateral ventricles. Single unit

responses were strongest in cortex and weakest in thalamus. Our fMRI findings indicated that several brain regions, including somatosensory/motor cortices, hippocampus, and thalamus showed BOLD responses to TES. However, brain regions around the ventricles even in deep structures were also affected by TES. Our results indicate that TES can affect deeper brain regions via current shunting of the ventricles. We are also planning to further confirm these results by quantifying the c-Fos expression induced by TES.

Disclosures: **M. Voroslakos:** None. **T.M. Arefin:** None. **J. Zhang:** None. **L. Alon:** None. **D.K. Sodickson:** None. **G. Buzsaki:** None.

Poster

PSTR181. Neural Stimulation to Understand Brain Function and Dysfunction

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR181.14/WW12

Topic: I.08. Methods to Modulate Neural Activity

Support: UH2/UH3 NS95495
R01-NS092882
UH2/UH3-NS95495

Title: Brain Restoration-Intelligent-Sensing-Stimulation-Ecosystem (BrainRISE): Application for epilepsy

Authors: *V. KREMEN[1], V. SLADKY[1], F. MIVALT[1,4], K. MCQUOWN[4], T. RYLAARSDAM[4], B. JOSEPH[2], I. BALZEKAS[2], V. MARKS[5], L. WHEELER[5], N. KISSOON[1], B. KLASSEN[1], L. JACKSON[1], N. GREGG[1], B. H. BRINKMANN[2], J. VAN GOMPEL[2], K. MILLER[3], G. WORRELL[1];
[1]Neurol., [3]Neurosurg., [2]Mayo Clin., Rochester, MN; [4]Windy City Lab., Chicago, IL; [5]Mayo Clin. Grad. Sch. of Biomed. Sci., Rochester, MN

Abstract: Rationale: Drug resistant epilepsy is common, effecting millions of people worldwide. It often has a significant impact on quality of life beyond seizures, including cognitive, sleep, and behavioral changes. Neuromodulation is an effective treatment, but has primarily focused on treating seizures and much less is known about the impact on comorbidities. Moreover, current mobile devices for recording human physiology and behavior that are useful for fully characterizing seizures and comorbidities are largely limited to wearable devices that lack robust, reliable protocols to easily connect and precisely synchronize data streams from multiple devices and human behavior (e.g., motor, mood, memory). Methods: The BrainRISE ecosystem was created for tracking epilepsy and comorbidities by integrating implantable neural sensing and stimulation devices, smartphones, and wearable sensors with local and cloud computing. BrainRISE is shown to enable precise millisecond synchronization of multimodal physiologic signals, stimuli, and human behavior in natural and virtual environments. This allows physicians to collect large-scale, multimodal data that can be used to develop algorithms and adaptively

optimize therapy. Results: BrainRISE was shown to be safe in pre-clinical testing in canines and has received an Investigational Device Exemption from the FDA for an early feasibility study in 20 patients with epilepsy. Currently, 10 patients with epilepsy have been using the system over multiple months to track seizures, sleep, memory, and mood. The impact of low frequency and high frequency electrical stimulation on sleep, memory and mood comorbidities was investigated, and the preliminary data support the potential benefits of low frequency neuromodulation for sleep and memory function. Our analysis of sleep patterns revealed that electrical brain stimulation of anterior nucleus of thalamus (EBS-ANT) influences sleep in all five patients, with high frequency EBS-ANT disrupting sleep by increasing wakefulness after sleep onset and decreasing both NREM and REM sleep duration (on average 6.47 ± 0.9 hours). In contrast, low frequency EBS-ANT preserved overall sleep duration and sleep architecture in all five patients, including NREM and REM sleep (on average 8.2 ± 0.5 hours). Conclusion: BrainRISE can accelerate discovery and therapy evaluation, and enable fundamental neuroscience investigations in ambulatory humans in natural and virtual environments.

Disclosures: **V. Kremen:** F. Consulting Fees (e.g., advisory boards); Certicon a.s.. **V. Sladky:** None. **F. Mivalt:** None. **K. McQuown:** None. **T. Rylaarsdam:** None. **B. Joseph:** None. **I. Balzekas:** None. **V. Marks:** None. **L. Wheeler:** None. **N. Kissoon:** None. **B. Klassen:** None. **L. Jackson:** None. **N. gregg:** None. **B.H. Brinkmann:** E. Ownership Interest (stock, stock options, royalty, receipt of intellectual property rights/patent holder, excluding diversified mutual funds); Cadence Neuroscience Inc. **J. Van Gompel:** E. Ownership Interest (stock, stock options, royalty, receipt of intellectual property rights/patent holder, excluding diversified mutual funds); Cadence Neuroscience Inc.. **K. Miller:** None. **G. Worrell:** E. Ownership Interest (stock, stock options, royalty, receipt of intellectual property rights/patent holder, excluding diversified mutual funds); Cadence Neuroscience Inc.

Poster

PSTR181. Neural Stimulation to Understand Brain Function and Dysfunction

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR181.15/WW13

Topic: I.08. Methods to Modulate Neural Activity

Support: NIH UH3NS095495

Title: Depression and anxiety in temporal lobe epilepsy show multiday cycles and associations with epileptiform brain activity

Authors: *I. BALZEKAS[1], T. J. RICHNER[2], V. SLADKY[1], F. MIVALT[2], N. GREGG[2], J. TRZASKO[1], P. E. CROARKIN[1], V. KREMEN[1], G. WORRELL[1];
[2]Neurol., [1]Mayo Clin., Rochester, MN

Abstract: *Background*: Although bidirectional relationships between depression and anxiety and epilepsy have been described, the relationship between depression and anxiety symptoms and

inter-ictal epileptiform discharges (IEDs) and seizures remains unclear. Comparisons of psychiatric symptoms, seizures, and IEDs have had inadequate long-term intracranial electroencephalographic (iEEG) recordings and statistical challenges in interpreting irregularly sampled psychiatric data. *Objective*: We integrated chronic, ambulatory iEEG monitoring and long-term ecological momentary assessments (EMAs) to quantify associations between seizures, IEDs, and depression and anxiety symptoms. We developed and implemented a method for cycle identification in sparsely and irregularly sampled neuro-behavioral data to detect multiday cycles of depression and anxiety symptoms. *Methods*: Three patients with temporal lobe epilepsy (TLE) participated under the Brain Initiative project "Neurophysiologically Based Brain State Tracking & Modulation in Focal Epilepsy (FDA IDE G180224)". Participants were implanted with the investigational Medtronic Summit RC+S ™ device. Continuous iEEG signals were recorded from bilateral hippocampi of ambulatory participants in their home environment. Seizures and IED rates were extracted from the iEEG recordings. Participants randomly completed an EMA describing depression and anxiety symptoms 1-4 times per week. Multiday cycles were identified in the EMA timeseries via basis pursuit denoising with polynomial detrending (BPWP). *Results*: We collected 1-2 years of iEEG and EMAs in each participant. We identified patient-specific, multiday cycles of EMA score around 100 days in all three participants. The average phase of high and low EMA scores differed significantly within patient-specific, underlying, multiday, IED rate cycles (S1:$p=0.0003$, S2:$p<0.0001$, S3:$p=0.0197$;Watson-Williams test). The most severe anxiety and depression symptoms followed periods of increased IEDs and seizure risk. *Conclusions:* We successfully integrated neural and psychiatric data to explore long-term neurobehavioral dynamics in TLE. This is the first quantitative demonstration of associations between multiday cycles of IEDs, seizures, and depression and anxiety symptoms in TLE. We demonstrated that BPWP is a viable method to identify cycles in sparsely and irregularly sampled timeseries. We anticipate this will be a powerful tool to evaluate cycles of behavior in other neuropsychiatric processes.

Disclosures: I. Balzekas: None. **T.J. Richner:** None. **V. Sladky:** None. **F. Mivalt:** None. **N. gregg:** None. **J. Trzasko:** None. **P.E. Croarkin:** None. **V. Kremen:** None. **G. Worrell:** None.

Poster

PSTR181. Neural Stimulation to Understand Brain Function and Dysfunction

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR181.16/WW14

Topic: I.08. Methods to Modulate Neural Activity

Support: NIH Grant UH2/UH3-NS95495
NIH Grant R01-NS092882
Mayo Clinic RFA CCaTS-CBD Pilot Awards for Team Science 2023
Investigational Medtronic Summit RC+S (TM) devices

Title: Very slow seizure-related impedance changes: a pilot study in humans with drug-resistant temporal lob epilepsy (TLE)

Authors: *J. CUI[1], F. MIVALT[2], V. SLADKY[2], N. M. GREGG[1], B. N. LUNDSTROM[2], E. K. ST. LOUIS[2], B. H. BRINKMANN[1], J. J. VAN GOMPEL[1], K. J. MILLER[3], V. KREMEN[1], G. A. WORRELL[1];
[1]Dept. of Neurol. and Dept. of Physiol. and Biomed. Engin., [2]Dept. of Neurol., [3]Dept. of Neurologic Surgery, Mayo Clin., Rochester, MN

Abstract: *Rationale* Fast impedance changes, on millisecond timescales due to opening of ion channels in active neuron membrane, and slow impedance change, occurring over seconds to minutes attributed to cell swelling, have been observed during seizures. However, little is known about the very slow impedance changes occurring on timescales of minutes to hours. This knowledge may provide insights into the mechanism of seizure generation and be useful for seizure forecasting. In this investigation, we studied seizure-related impedance change over 24 hours in humans with TLE. *Methods* Five peoples with drug resistant TLE were implanted with bilateral electrodes in the thalamus, amygdala, and hippocampus. One patient was excluded from this analysis due to a prior epilepsy surgery resulting in inconsistent seizure-related impedance change. The impedance was sampled nonuniformly (every 5 - 15 minutes) over multiple months. Seizures were identified from both hemispheres, but mainly from the more epileptogenic left side. The distributions of seizure onset times were quantified with circular histograms. Surrogate data segments from days without seizures were generated and aligned to the same distributions of seizure onset as the data segments with seizures. Cross-correlation between impedance and seizures was estimated and compared to cross-correlation with surrogate seizures. *Results* The estimated cross-correlation showed a peak in correlation in the amygdala-hippocampus and posterior hippocampus. In left thalamus, the mean value of normalized impedance related to real seizures was lower than that related to surrogate seizures about 0-10 hours before seizure onset. In left amygdala-hippocampus, the mean value of normalized impedance related to real seizures was higher than that related to surrogate ones about 0-10 hours before seizure onset. Significant difference was found in the left side of posterior hippocampus. The normalized impedance related to real seizures was significantly higher (mean values were separated by > 2 SEM) than that related to the surrogate seizures ~0-4 hours before seizure onset. *Conclusion* The peaks of cross-correlated impedance may suggest that seizure onsets could be phase locked to the circadian cycle of impedance. The impedance of thalamus is likely lowered, while that of hippocampus elevated hours before the seizure onset. The significant increase of impedance 0-4 hours before seizure onset in the left posterior hippocampus is potentially useful for forecasting seizure alert.

Disclosures: J. Cui: None. **F. Mivalt:** None. **V. Sladky:** None. **N.M. Gregg:** None. **B.N. Lundstrom:** B. Contracted Research/Research Grant (principal investigator for a drug study, collaborator or consultant and pending and current grants). If you are a PI for a drug study, report that research relationship even if those funds come to an institution.; Cadence Neuroscience Inc. C. Other Research Support (receipt of drugs, supplies, equipment or other in-kind support); Medtronic Inc. **E.K. St. Louis:** None. **B.H. Brinkmann:** B. Contracted Research/Research Grant (principal investigator for a drug study, collaborator or consultant and pending and current grants). If you are a PI for a drug study, report that research relationship even if those funds come to an institution.; Cadence Neuroscience Inc. C. Other Research Support (receipt of drugs, supplies, equipment or other in-kind support); Medtronic Inc. **J.J. Van Gompel:** B. Contracted Research/Research Grant (principal investigator for a drug study, collaborator or consultant and pending and current grants). If you are a PI for a drug study, report that research relationship

even if those funds come to an institution.; Cadence Neuroscience Inc. C. Other Research Support (receipt of drugs, supplies, equipment or other in-kind support); Medtronic Inc. **K.J. Miller:** None. **V. Kremen:** None. **G.A. Worrell:** B. Contracted Research/Research Grant (principal investigator for a drug study, collaborator or consultant and pending and current grants). If you are a PI for a drug study, report that research relationship even if those funds come to an institution.; Cadence Neuroscience Inc. C. Other Research Support (receipt of drugs, supplies, equipment or other in-kind support); Medtronic Inc.

Poster

PSTR181. Neural Stimulation to Understand Brain Function and Dysfunction

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR181.17/WW15

Topic: I.08. Methods to Modulate Neural Activity

Title: Predictive causal modeling of evoked intracranial EEG response to medial temporal lobe stimulation in patients with epilepsy

Authors: *G. ACHARYA[1], K. A. DAVIS[2], E. NOZARI[1];
[1]Univ. of California, Riverside, Riverside, CA; [2]Neurol., Hosp. of the Univ. of Pennsylvania, Philadelphia, PA

Abstract: Closed-loop neurostimulation has gained significant attention as a potential treatment for drug-resistant epilepsy (DRE). However, due to the intricate nature of the brain and the mechanistic complexity of DRE, current clinical approaches rely on rigid and manual adjustments of parameters. Meanwhile, the realization of fully responsive seizure control algorithms that determine stimulation timing and parameters in closed loop has been limited by the absence of in silico models that precisely capture the dynamic network response of the brain to neurostimulation. In our work, we take a first step towards addressing this gap by data-driven neurodynamical predictive modeling of large-scale neurostimulation-evoked intracranial EEG (iEEG) activity in human subjects.Using parametrically rich iEEG recordings from the Restoring Active Memory (RAM) project, we trained and compared several subject-channel-specific state-of-the-art dynamical system models, including various linear and nonlinear autoregressive (AR) models, long short-term memory networks, and sparse identification methods. Despite significant heterogeneity among subjects (n = 10) and even same-subject channels in the structure of the models with the highest predictive power, on average, evoked iEEG dynamics were best explained by switched linear AR models (in > 80% channels). We found about 300ms of causal historical dependence in iEEG, with values from more distant pasts leading to only marginal improvements in the overall prediction accuracy. Furthermore, we observed a consistently distance-dependent pattern of how stimulation affects the iEEG response in different channels. The response at the stimulation site and at its close vicinity (< 20mm) was heavily dominated by direct stimulation effects and thus received little or no causal effect from other brain regions. The strength of network interactions gradually intensified with increasing distance from the anode, reached its peak at an intermediary distance of around 70mm (±23mm), and

decayed thereafter. Nodes farthest from the anode showed little to no causal effect from either the stimulation or other channels, consistent with prior work in resting-state iEEG. Additionally, we found switched-linear models able to generalize across different stimulation amplitudes and frequencies, including STIM OFF durations, but not across different subjects or even different sessions within the same subject. This provides a significant opportunity for using abundant STIM OFF data to augment evoked iEEG in predictive modeling while reinforcing the need for adaptive tuning of models to mitigate nonstationarities in iEEG dynamics.

Disclosures: G. Acharya: None. **K.A. Davis:** None. **E. Nozari:** None.

Poster

PSTR181. Neural Stimulation to Understand Brain Function and Dysfunction

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR181.18/WW16

Topic: I.08. Methods to Modulate Neural Activity

Support: NINDS grant DP1NS122038
NINDS grant R01NS125137

Title: Non-epileptiform background features demonstrate multidien cycles in long-term EEG device recordings

Authors: *W. OJEMANN, B. SCHEID, S. MOUCHTARIS, A. LUCAS, J. LAROCQUE, C. AGUILA, L. CACIAGLI, E. CONRAD, B. LITT;
Bioengineering, Univ. of Pennsylvania, Philadelphia, PA

Abstract: Recent research leveraging longitudinal EEG recordings reports patient-specific, multi-day cycles in device-detected events that coincide with increased likelihood of clinical seizures, but their physiological mechanism is poorly understood. Understanding these cycles could elucidate mechanisms generating seizures and advance drug and neurostimulation therapies. We analyzed regularly scheduled data epochs from 23 patients implanted with the NeuroPace RNS device over periods of 2-6 years to explore the relationship between cycles in detected events (dIEA) (A), interictal spikes, background EEG features and neurostimulation. From interictal recordings with and without stimulation we calculated band-limited band power and connectivity in four different frequency bands: theta (4-8Hz) alpha (8-13Hz), beta (13-30Hz), and gamma (30-100Hz). We then built multivariate SVM models to relate these features to dIEA cycle phase (B). Background EEG features tracked the cycle phase of dIEA in all patients (AUC: 0.63 [0.56 - 0.67]) with significantly higher effect size compared to clinically annotated spike rate alone (AUC: 0.55 [0.53-0.61], ranksum $p<0.01$). We observed significant population trends in elevated theta and alpha band power and theta and beta connectivity at the cycle peaks (sign test, $p < 0.05$) (E). Variance in the specific EEG feature that best tracked dIEA cycle phase (C,D) suggests a patient-specific effect. In the period directly after stimulation we observed a decreased association between cycle phase and EEG features compared to

background recordings (AUC: 0.58 [0.55-.64]) (F) largely driven by patients with neocortical implants. Our findings suggest that seizure-correlated cycles are associated with background measures of brain state most strongly in the neocortex, and that neurostimulation disrupts this relationship. These results may help elucidate mechanisms underlying seizure generation, provide new biomarkers for seizure risk, and facilitate monitoring, treating and managing epilepsy with implantable devices.

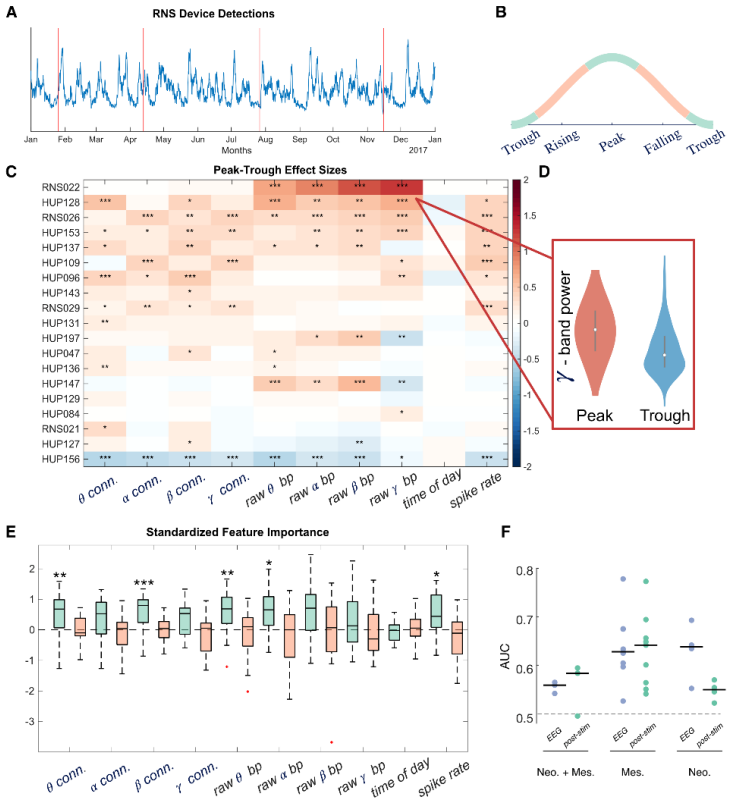


Figure 1. A) RNS device detection signal normalized in between detection parameter visits (orange lines). Seizures preferentially occur at the rising phase of this signal. B) Phases of the dIEA signal. We compared peak vs. trough (green) and rising vs. falling phases (orange). C) Effect sizes (Cohen's d) comparing feature values at signal peaks vs troughs (*** p<0.005, **p < 0.01, *p < 0.05). D) Example feature distribution comparing gamma band power between the peaks and troughs of the dIEA signal. E) Population distribution of multivariate model coefficients showing the population trend of higher band power and connectivity at the peaks of dIEA cycles compared to the troughs. F) Comparison of AUC distributions between background EEG and post-stim models in patients with different implant depths: neocortical, mesial temporal, and both.

Disclosures: W. Ojemann: None. **B. Scheid:** None. **S. Mouchtaris:** None. **A. Lucas:** None. **J. LaRocque:** None. **C. Aguila:** None. **L. Caciagli:** None. **E. Conrad:** None. **B. Litt:** None.

Poster

PSTR181. Neural Stimulation to Understand Brain Function and Dysfunction

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR181.19/WW33

Topic: I.08. Methods to Modulate Neural Activity

Support: R01-NS092882
Cadence Neuroscience Inc.

Title: A Canine with Naturally Occurring Generalized Epilepsy: Thalamus Local Field Potentials and Electrical Stimulation

Authors: *F. MIVALT[1], N. GREGG[1], I. KIM[1], K. MILLER[1], J. VAN GOMPEL[1], S.-Y. CHANG[1], B. H. BRINKMANN[1], V. KREMEN[1], W. SHEFFIELD[2], G. WORRELL[1];
[1]Mayo Clin., Rochester, MN; [2]Cadence Neurosci. Inc., Redmond, WA

Abstract: Rationale: Epilepsy is a common, naturally occurring neurological disorder in dogs and shares common features with human epilepsy. Dogs can accommodate devices designed for humans, making them a valuable translational platform for the development of novel implantable neural sensing and recording (INSR) devices. The functionality of next-generation INSR devices enables streaming local field potential (LFP) data, electrical brain stimulation (EBS) and tracking of electrophysiological biomarkers and behavior in natural environments.
Methods: A female hound dog with nocturnal generalized convulsive seizures and interictal generalized polyspike and wave epileptiform activity recorded from scalp EEG was permanently implanted with 4 leads (16 total contacts) targeting bilateral dorsal-medial nucleus of the thalamus (DM), centromedian nucleus of the thalamus (CM) and pulvinar nucleus of the thalamus (Pul) using the investigational Cadence Neuroscience implantable INSR device. Brainlab neuronavigation stereotactic system and 0.8 mm isometric MPRAGE and FGATIR MRI were used for targeting. Thalamic recordings were sufficient to use for sleep state differentiation using previously developed techniques.
Results: Intraoperative recording and electrical stimulation revealed generalized bilateral interictal epileptiform spikes and evoked response potentials. During anesthesia CM EBS (100 Hz, 90 us, 1.5 mA) was able to repeatedly used to reliable arouse the dog from general 1% isofluorine anesthesia. Arousals were coupled with clear behavior manifestations and intracranial LFP changes. Chronic intracranial LFP recordings post-surgery revealed interictal epileptiform spikes manifesting dominantly during non-REM compared to awake (non-REM: 59.36 ± 56.83 vs Awake: 1.21 ± 3.78 spikes per 10 mins, $p < 0.001$) with $5.94\ \% \pm 10.64\ \%$ of spikes being bilateral. The highest recorded spike count was in CM 93.67 ± 60.09 compared to other targets 37.92 ± 42.92 spikes per 10 mins ($p < 0.01$).
Conclusions: The genetic generalized epilepsies (GGE) are a broad group of human epilepsies with generalized seizures and LFP correlates that may respond to EBS. Naturally occurring animals with GGE provide a platform for developing new EBS therapies. We show that a novel 4-lead chronic sensing-and-stimulation device enables wake/sleep state classification, and

interictal epileptiform discharge quantification, and that CM stimulation modulates level of arousal. Future efforts will assess the therapeutic impact of thalamic stimulation on canine generalized epilepsy. Next generation devices provide new methods to characterize and modulate epilepsy and behavior.

Disclosures: **F. Mivalt:** B. Contracted Research/Research Grant (principal investigator for a drug study, collaborator or consultant and pending and current grants). If you are a PI for a drug study, report that research relationship even if those funds come to an institution.; Cadence Neuroscience Inc.. **N. gregg:** None. **I. Kim:** None. **K. Miller:** None. **J. Van Gompel:** E. Ownership Interest (stock, stock options, royalty, receipt of intellectual property rights/patent holder, excluding diversified mutual funds); Cadence Neuroscience Inc.. **S. Chang:** None. **B.H. Brinkmann:** E. Ownership Interest (stock, stock options, royalty, receipt of intellectual property rights/patent holder, excluding diversified mutual funds); Cadence Neuroscience Inc.. **V. Kremen:** None. **W. Sheffield:** A. Employment/Salary (full or part-time); Cadence Neuroscience Inc.. E. Ownership Interest (stock, stock options, royalty, receipt of intellectual property rights/patent holder, excluding diversified mutual funds); Cadence Neuroscience Inc. **G. Worrell:** E. Ownership Interest (stock, stock options, royalty, receipt of intellectual property rights/patent holder, excluding diversified mutual funds); Cadence Neuroscience Inc.

Poster

PSTR181. Neural Stimulation to Understand Brain Function and Dysfunction

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR181.20/WW34

Topic: I.08. Methods to Modulate Neural Activity

Support: NIH Grant T32GM007739
HDRF Grant 200192-01

Title: Investigating the Effects of rTMS Treatment on Resting State Functional Connectivity in Major Depressive Disorder

Authors: *A. N. DUA[1], K. DUNLOP[2], C. LYNCH[1], D. M. BLUMBERGER[2], J. DOWNER[2], L. GROSENICK[1], C. LISTON[1];
[1]Weill Cornell Med., New York, NY; [2]Univ. of Toronto, Toronto, ON, Canada

Abstract: Non-invasive stimulation therapies such as repetitive transcranial magnetic stimulation (rTMS) are effective antidepressant treatments for a subset of patients with treatment resistant major depressive disorder (MDD). The mechanisms through which rTMS produces an antidepressant response remain poorly understood at both the molecular and circuit levels. Neuroimaging methods such as resting state functional MRI (rs-fMRI) allow us to investigate activity patterns in the brain using measures like resting state functional connectivity (rsFC). In depression, pre-treatment rsFC features have been shown to accurately predict disease status, disease biotypes, and treatment response to both drug therapies and rTMS. To better understand

the circuit-level mechanism of rTMS treatment in depression, we used pre- and post-treatment rs-fMRI scans of over 250 MDD patients to investigate the impact of rTMS treatment on rsFC and other rs-fMRI-derived measures. The effect of rTMS treatment on these measures were assessed across multiple scales of brain organization (regional, subnetwork and network organization) in both whole brain and pre-selected, MDD-associated networks of interest. Through these analyses we find that most rsFC features are remarkably stable over time. Our work provides an expansive description of the circuit-level effect of rTMS in MDD.

Disclosures: A.N. Dua: None. **K. Dunlop:** None. **C. Lynch:** None. **D.M. Blumberger:** None. **J. Downer:** None. **L. Grosenick:** None. **C. Liston:** None.

Poster

PSTR181. Neural Stimulation to Understand Brain Function and Dysfunction

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR181.21/WW35

Topic: I.08. Methods to Modulate Neural Activity

Support: Tiny Blue Dot Foundation
NIH grant K24-NS088568
CURE Epilepsy Taking Flight award (RZ)

Title: Frontal disconnection during lack of arousability in humans

Authors: *R. ZELMANN[1], A. C. PAULK[1], F. TIAN[2], G. A. BALANZA VILLEGAS[2], J. DEZHA PERALTA[1], G. COSGROVE[5], Z. WILLIAMS[3], R. RICHARDSON[3], D. D. DOUGHERTY[4], P. L. PURDON[2], S. S. CASH[1];
[1]Neurol., [3]Neurosurg., [4]Psychiatry, [2]Massachusetts Gen. Hosp., Boston, MA; [5]Neurosurg., Brigham and Women's Hosp., Boston, MA

Abstract: What happens in the human brain when we are unconscious and unarousable? Theoretical approaches and experimental studies differ concerning which brain regions are necessary for consciousness and arousability. Direct experimental evidence to resolve this debate requires identifying the global, network, and regional involvement during wake vs. arousable unconsciousness (sleep) vs. non-arousable unconsciousness (propofol-induced general anesthesia).
In 20 patients with depth electrodes implanted to determine the origin of their epileptic seizures, we delivered pseudo-random multi-site bipolar single pulse electrical stimulation while simultaneously recording intracranial EEG (115 stimulation sites, 2357 recording channels). We estimated complexity, connectivity features, and inter-trial variability. We compared non-REM sleep vs. awake (N=13, s=99 stimulated channels), anesthesia during electrode removal vs. awake in the operating room (N=14; s=36), and awake in different environments (N=13; s=34). Cortico-cortical evoked potentials (CCEP) were overall detected in 44% of the channels. Perturbational complexity, connectivity, and the response's amplitude were significantly

reduced, while inter-stimulation trial variability increased during both unconscious states, compared to wake (p<<0001). These changes were more pronounced during anesthesia than sleep. They also involved different cortical engagement. During sleep, relative changes, normalized by awake in each environment, were mostly uniformly distributed across the brain. During anesthesia, the prefrontal cortex was the most disrupted (Fig. 1).
These findings provide direct evidence of the different neural signatures in humans for loss of consciousness and loss of arousability. The lack of arousability during anesthesia arises from altered overall physiology and from a disconnection between the prefrontal and other brain areas.

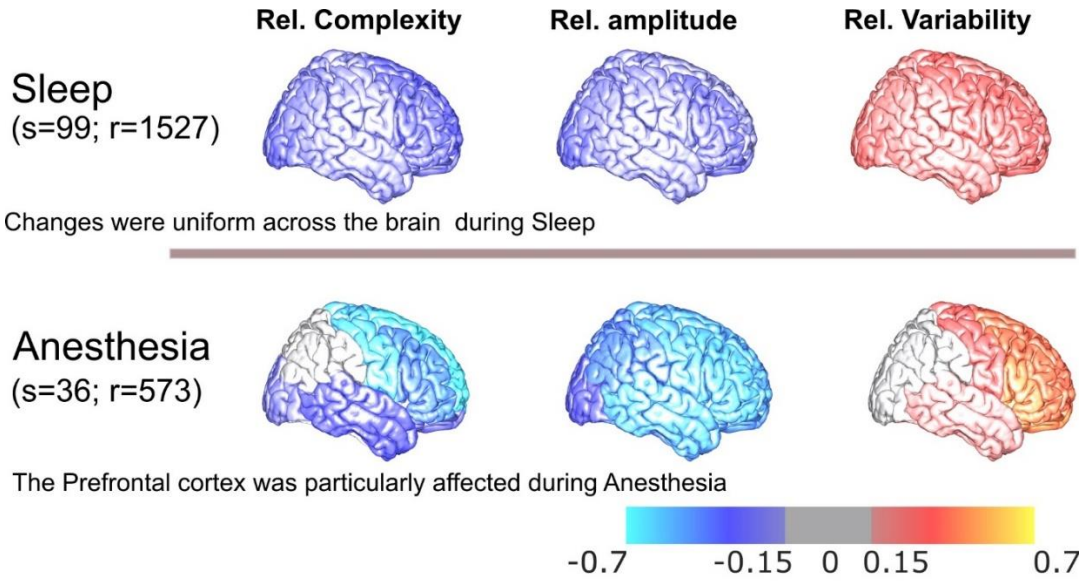


Figure 1. Relative perturbational complexity (PCI^{st}), amplitude, and variability (standard deviation) during Sleep and Anesthesia, normalized by wake in the same environment. The anatomical distribution of relative complexity and amplitude following stimulation showed a uniform decrease across the brain during natural sleep, and a predominant decrease in frontal regions during propofol induced general anesthesia. Similarly, variability increased uniformly across the brain during sleep and predominantely in frontal regions during anesthesia.

Disclosures: **R. Zelmann:** None. **A.C. Paulk:** None. **F. Tian:** None. **G.A. Balanza Villegas:** None. **J. Dezha Peralta:** None. **G. Cosgrove:** None. **Z. Williams:** None. **R. Richardson:** None. **D.D. Dougherty:** C. Other Research Support (receipt of drugs, supplies, equipment or other in-kind support); Medtronic. S.S.C.. E. Ownership Interest (stock, stock options, royalty, receipt of intellectual property rights/patent holder, excluding diversified mutual funds); Innercosmos and Neurable, MGH patents. F. Consulting Fees (e.g., advisory boards); Sage and Celanese. **P.L. Purdon:** E. Ownership Interest (stock, stock options, royalty, receipt of intellectual property rights/patent holder, excluding diversified mutual funds); PASCALL Systems, Inc., patent licensed to Masimo. **S.S. Cash:** F. Consulting Fees (e.g., advisory boards); Beacon Biosignals.

Poster

PSTR181. Neural Stimulation to Understand Brain Function and Dysfunction

Location: WCC Halls A-C

Time: Sunday, November 12, 2023, 1:00 PM - 5:00 PM

Program #/Poster #: PSTR181.22/WW36

Topic: I.08. Methods to Modulate Neural Activity

Support: CURE Epilepsy Taking Flight award
R01NS062092
NIH grant K24-NS088568

Title: A deep learning algorithm for the separation of physiological and pathological intracranial electroencephalogram data in epileptic patients

Authors: *J. DEZHA PERALTA[1], M. FAN[1], A. PAULK[2], S. S. CASH[1], R. ZELMANN[1]; [1]Neurol., Massachusetts Gen. Hosp., Boston, MA; [2]Neurol., Dept. of Neurology, Massachusetts Gen. Hosp., Boston, MA

Abstract: A deep learning algorithm for the separation of physiological and pathological intracranial electroencephalogram data in epileptic patients
Authors: Jaquelin Dezha Peralta, Miaolin Fan, Angelique C. Paulk, Sydney S. Cash, and Rina Zelmann
AbstractIntracranial EEG data provide a unique window into the human brain. Opportunities to acquire such data arise from patients implanted with electrodes to characterize their epileptic seizure, therefore, affecting the interpretation of cognitive data. This dilemma could be bypassed by separating the physiological and pathological EEG, although this is challenging. Here, we used a deep learning algorithm to detect and discard channels classified as epileptic and then to detect normal brain responses to single pulse electrical stimulation (SPES). In 9 epileptic patients implanted with intracranial electrodes, we delivered SPES at various sites while recording the stimulation responses. We included 84 stimulation channels (patient mean = 9.3, range [3-14]) and 1349 recording channels (mean = 149.9 [114-204]) with 137 channels (mean = 15.2 [2-33]) in the Seizure Onset Zone (SOZ). A one-second response (recorded at 2 kHz, stimulation artifact removed, filtered between 0.3 Hz and 100 Hz, and downsampled to 512 Hz) was the input to a Convolutional Neural Network (CNN) to classify the recording channels into two classes: a channel in the SOZ or outside the SOZ. We then classified the recording channels into three classes: a channel in the SOZ (pathological), a channel outside of the SOZ with a Cortico-Cortical-Evoked-Potential (CCEP) response (physiological), or a channel outside of the SOZ with no response (physiological). We modified 1D-CNN (https://github.com/geekfeiw/Multi-Scale-1D-ResNet) for single-channel inputs, with a weighted cross-entropy loss function, and validated with a 5-fold cross-validation. The 2-class model classified recording channels as SOZ vs. outside with a mean 82% accuracy (range [75 - 88%]), 43% sensitivity [21- 66%], and 88% specificity [82- 93%]. The 3-class model classified the recording channels in the SOZ, channels outside the SOZ with a CCEP response, and channels outside the SOZ with no response with a mean 55% accuracy (range [40 - 74%]). We trained the 3-class model with shuffled data which produced a 34% accuracy and a T-test of the 3-class model ($p<<0.0001$) support the viability of the 3-class model. This CNN can classify SPES responses to separate pathological and physiological EEG in epileptic patients and detect desired CCEP responses. The ability to select the type of EEG desired can amplify the impact of data collected from epileptic patients.

Disclosures: **J. Dezha Peralta:** None. **M. Fan:** None. **A. Paulk:** None. **S.S. Cash:** F. Consulting Fees (e.g., advisory boards); Beacon Biosignals. **R. Zelmann:** None.