REVIEW

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Celiac Disease: A Disorder Emerging from Antiquity, Its Evolving Classification and Risk, and Potential New Treatment Paradigms

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Celiac disease is a chronic genetically based gluten-sensitive immune-mediated enteropathic process primarily affecting the small intestinal mucosa. The disorder classically presents with diarrhea and weight loss; however, more recently, it has been characterized by subclinical occult or latent disease associated with few or no intestinal symptoms. Diagnosis depends on the detection of typical histopathological biopsy changes followed by a gluten-free diet response. A broad range of clinical disorders may mimic celiac disease, along with a wide range of drugs and other therapeutic agents. Recent and intriguing archeological data, largely from the Gobleki Tepe region of the Fertile Crescent, indicate that celiac disease probably emerged as humans transitioned from hunter-gatherer groups to societies dependent on agriculture to secure a stable food supply. Longitudinal studies performed over several decades have suggested that changes in the prevalence of the disease, even apparent epidemic disease, may be due to superimposed or novel environmental factors that may precipitate its appearance. Recent therapeutic approaches are being explored that may supplement, rather than replace, gluten-free diet therapy and permit more nutritional options for future management. **(Gut Liver 2015;9:28-37)**

Key Words: Celiac disease; Celiac disease history; Occult and latent celiac disease; Sprue-like intestinal disease; Celiac disease therapy

INTRODUCTION

Celiac disease is a life-long gluten-sensitive immune-mediated disorder affecting the small intestinal mucosa. Reviews have recently appeared focused on prevalence, diagnosis, pathogenesis and treatment.[1,2] The disorder is thought to be restricted to genetically-susceptible individuals, and has been likened to an "iceberg disease," since subclinical presentations with few or no intestinal symptoms are becoming more readily recognized. Common or classical features include diarrhea and weight loss, but celiac disease is now often first detected in those presenting with a wide array of clinical disorders such as iron deficiency anemia, osteoporosis, "autoimmune" conditions, like dermatitis herpetiformis or autoimmune thyroiditis, and even some neurological disorders, including dementia.[3,4] This variability in the initial clinical presentation appears largely related to genetic and immunological factors, age of onset, extent and degree of small intestinal mucosal inflammation, gender, and familial nature.[5] Finally, and particularly in recent years, celiac disease has appeared in an epidemic pattern,[6] possibly related to age of introduction of dietary gluten, specific infections, medication use and supplements.[7]

CRITICAL ELEMENTS IN DIAGNOSIS

Diagnosis, particularly in adults, depends on an initial small intestinal biopsy that shows characteristic or typical pathological features of untreated celiac disease. As the disease is a gluten-dependent disorder, improvement on a gluten-free diet is also critically essential.[3] Biopsy of the small intestine has limitations, particularly if poorly oriented specimens are submitted in fixative by the clinician for laboratory processing, sometimes leading to tangential sectioning and very difficult biopsy interpretation by even expert pathologists. Predisposing genetic factors, including human leukocyte antigen markers, HLA DQ2 and HLA DQ8, may be present and serological changes, including antibodies to tissue transglutaminase antigen (tTG), are usually evident.

Serological studies have also often been employed as a screening tool for celiac disease, particularly for population

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studies, or for clinical case finding since biopsy changes are usually present with a positive serological test. However, the presence of tTG antibodies alone is not definitive for diagnosis of celiac disease. False negative tests may result (particularly with immunoglobulin deficiency syndromes, i.e., IgA deficiency) and even strongly positive tTG tests without biopsy evidence for celiac disease have been reported.[8] Quantitative tTG assessment may also be helpful since there may be a correlation with the degree of biopsy change and some, but not all, believe that reduced tTG titers on a gluten-free diet may be reflective of clinical improvement and permit assessment of diet compliance. In recent years, guidelines have also appeared suggesting a diagnostic approach.[9] A biopsy showing the typical pathological features of celiac disease followed by a clear response to a gluten-free diet is essential. Empirical use of a gluten-free diet without an initial biopsy, even if believed to be symptomatically helpful, is not recommended.

EMERGING HISTORICAL ASPECTS

Some of the most intriguing information has emerged in recent years owing to archeological studies. Wheat cultivation methods first appeared in the Fertile Crescent about 10,000 years ago. Celiac disease may have developed as a distinct disorder with the transition of hunter-gatherer groups into human workforces capable of agriculture. This “Neolithic revolution”[10] is believed to have permitted competitive survival over other hunter-gatherer groups owing to more secure food supplies. Over time, celiac disease has emerged as a major clinical disorder, currently thought on the basis of serological studies to affect up to about 2% of most genetically-predisposed human populations.[11]

The Gobleki Tepe in southeastern Turkey is now recognized as one of the most important modern archeological discoveries impacting on history and development of humans, and disorders, like celiac disease.[12] Here, recent excavations led not only to important information on origins of highly complex and ritualized societies, but also, as the “cradle of agriculture,” an appreciation for a high concentration for several wild forms of early domesticated plant species with an overlapping distribution, including wild forms of Einkorn and Emmer wheat, barley and other Neolithic founder crops. Later DNA fingerprinting studies also established a clear relationship with this wild Einkorn wheat species and other modern forms of grains that have evolved with increased and more immunogenic gliadin content.[13]

In 1856, Francis Adams[14] delivered a lecture to the Sydenham Society (based on translation of a Greek dialect) providing an account of the clinical features of celiac disease along with recommendations for treatment by a Greek physician, Aretaeus of Cappadocia, in the second century AD. Aretaeus used the word, “coeliac,” derived from the Greek, “koiliakos” (meaning “abdominal”) to detail a case record of celiac disease including symptoms of diarrhea and malabsorption. Subsequently, another case from antiquity was later suspected by Italian investigators in a young woman from the first century AD in a region also apparently exposed to wheat cultivation methods.[15]

Much later, in 1888, Samuel Gee,[16] a physician working at the Children’s Hospital on Great Ormond Street in London, provided the first modern clinical description of celiac disease in children, noting that the disorder might occur at any age, and suggested that attention to diet might ultimately lead to a cure. He also recorded a child that improved with a diet of mussels, followed by relapse after the mussel season ended. In 1924, Haas[17] from the United States published positive results with a banana diet, a popular treatment for decades. Subsequently, Dicke and his colleagues from the Netherlands provided clinical and laboratory evidence for gluten-free diet therapy, based on starvation and re-feeding effects on growth of children during the Second World War as well as measured endpoints for small intestinal absorption, including calculated coefficients of fecal fat absorption.[18]

In 1954, Paulley[19] from the United Kingdom detailed pathological changes in the small intestine based on surgical specimens from patients with steatorrhea. Later technological developments made the small bowel more accessible for direct imaging and pathological evaluation. In most recent years, new data has emerged on virtually every aspect of celiac disease, including potentially important and novel treatment options.[20]

Interestingly, even now, transition from food-gathering to food-producing societies continues. For example, some immune-mediated disorders, including celiac disease, have only recently been reported in the Coast Salish First Nations populations on the west coast of Canada.[21] These indigenous peoples were a culturally complex society that benefited from a temperate zone maritime climate, living in permanent villages of more than 1,000 residents with social stratification, including slaves and ranked nobility, multiple linguistic dialects and a distinctive art style. The Coast Salish lived largely on fish, fruits and berries without soil cultivation methods. Subsequently, potato and wheat cultivation methods were likely introduced through Russian, Spanish or British settlements from Alaska, California and eventually the Fraser River Valley, associated with the Hudson’s Bay Company. This has been hypothesized to have resulted in a rapid change in the environment, including the emergence of wheat cultivation methodologies, thought to be a critical element in development of celiac disease.[22]

EVOLVING CLINICAL AND PATHOLOGICAL SPECTRUM

Celiac disease presents as a spectrum of gluten-sensitive mucosal change, noted earlier in this journal, where representative photomicrographs can also be located.[23] This spectrum of celiac disease may be classified into a variety of clinical presentations.

Classical celiac disease is usually recognized in children or adults with diarrhea, weight loss and textbook clinical changes of malabsorption. In these, antibodies to tissue transglutaminase antigen are usually evident. Small bowel biopsies show typical changes with crypt hyperplasia and flattening of intestinal villi.

Occult or atypical celiac disease usually presents with limited or no intestinal symptoms. Extraintestinal features dominate and include changes such as iron deficiency anemia, fracture associated with osteopenia, peripheral neuropathy, infertility, abnormal liver chemistry tests, or skin rash characterized as dermatitis herpeformis. Subsequent evaluation reveals typical biopsy changes of untreated celiac disease, usually with positive serological studies.

Latent celiac disease may be defined by the presence of a predisposing gene, such as, HLA-DQ2 and/or HLA-DQ8, associated with architecturally-normal intestinal biopsies, sometimes with increased numbers of intraepithelial lymphocytes. Biopsy studies in dermatitis herpetiformis patients and no apparent changes of celiac disease showed some intriguing results.[24] In these, a high gluten-containing diet induced mucosal inflammatory changes in the small intestine typical of celiac disease while a gluten-free diet then caused resolution of intestinal symptoms and improvement in induced small intestinal mucosal morphological changes.[24] Similar results, using blinded biopsy specimens, in a patient with lymphoma and latent celiac disease were also noted.[25]

Refractory celiac disease usually occurs after age 50 years in already well documented celiac disease. In these, recurrent symptoms and biopsy changes occur despite strict diet adherence.[26,27] It is critical here for clinicians to note that if no response to a gluten-free diet has ever been demonstrated, celiac disease may not have ever been present.

"Sprue-like" enteropathy or unclassified sprue may be present.[26,27] This is not a refractory form of celiac disease, but represents an increasingly recognized broad clinical and pathological spectrum of possibly unrelated disorders that do not respond to a gluten-free diet. Some of these are noted in Tables 1 and 2.

Table 1. Sprue Syndromes

Disorder	Treatment
Celiac disease (classical, occult, latent)	Gluten-free diet
Oats-induced sprue-like small bowel disease	Restrict oats
Refractory celiac disease	Not known
Collagenous sprue (enteritis or enterocolitis)	Not known
Mesenteric lymph node cavitation syndrome	Not known
Other protein-induced mucosal disease (soy, milk)	Delete protein
Unclassified sprue (sprue-like intestinal disease)	Not known

All may cause diffuse severe ("flat") or moderate to severe changes in the mucosal architecture. Refractory celiac disease requires evidence of an initial gluten-free diet response. In unclassified sprue (sprue-like intestinal disease), no response to a gluten-free diet can be documented. Celiac disease has also been termed "celiac sprue" or "gluten-sensitive enteropathy." Adapted from Freeman HJ. Int J Celiac Dis 2014;2:6-10.[27]

Table 2. Sprue-Like Biopsy Changes

Disorder	Treatment
Infectious causes	
Specific agents (parasite, protozoan, mycobacteria)	Treat specific agent
Tropical sprue	Antibiotics and folic acid
Stasis syndrome (contaminated small bowel)	Antibiotics
Whipple's disease (or *Tropheryma whipplei*)	Antibiotics
Deficiencies	
Nutrients (zinc, vitamin B_{12}, folic acid)	Replace specific agent
Malnutrition (kwashiorkor)	Adequate dietary protein
Immune deficiency syndromes (transplant, HIV, common variable type, X-linked)	No specific treatment
Others	
Intestinal lymphangiectasia	Not known
Crohn's disease (with duodenal involvement)	No cause known
Postproctocolectomy enteropathy	No cause known
Graft-vs-host disease	Treat graft rejection
Immunoproliferative disease (lymphoma)	Often chemotherapy
Macroglobulinemia	Often chemotherapy
Zollinger-Ellison syndrome	Antisecretory treatment
Drug-induced small bowel disease	Remove drug
Microvillus inclusion disease	Not known

HIV, human immunodeficiency virus.

Several of these have only recently been described in either children or adults. For example, neonates with the onset of diarrhea days to months following birth have been discovered to have microvillus inclusion disease. Altered small intestinal mucosal structure occurs and histochemical staining with periodic acid-Schiff reagent typically suggests subapical inclusions in villus enterocytes.[28] Confirmatory ultrastructural studies reveal variable loss of the epithelial cell microvilli, microvillus inclusions and subapical vesicles. A specific mutation of myosin Vb (MYO5B) has been reported. Most recently, a form of variant microvillus inclusion disease has been described with a loss of syntaxin 3, an apical receptor involved in membrane fusion of apical vesicles in enterocytes.[29]

Moreover, similar "new" diseases have emerged at the adult end of the clinical spectrum. For example, a very distinctive sprue-like enteropathic process in the proximal small intestine has been recorded after colectomy for ulcerative colitis, including those treated with a pelvic pouch reconstruction procedure.[30,31] This entity appears to be uncommon, but is probably under-reported. Severe diarrhea and a marked nutritional deficiency may occur. Biopsies from the duodenum and proximal jejunum may be severely abnormal. Negative tTG antibodies have been noted and changes fail to respond to a gluten-free diet. To date, treatment has been largely empirical relying on significant immunosuppression combined with nutritional support.

Perhaps the most frequently recognized "new" forms of sprue-like enteropathy include "drug-induced" or "medication-related" forms of enteropathy that may cause severe diarrhea and nutrient malabsorption. Historically, these are not entirely novel, but the list is now expanding. Triparanol, for example, was an injected agent used over 50 years ago to induce an experimental animal model of celiac disease.[32] It was thought that this agent provoked labilization of lysosomal membranes within enterocytes leading to liberation of intracellular hydrolytic enzyme activities with resultant destruction of enterocytes. A syndrome in rats poisoned with this agent also appeared to respond to a gluten-free diet.

Table 3. Medications Causing Sprue-Like Small Bowel Disease

Medication	Example
Nonsteroidal anti-inflammatory drugs	Sulindac[34]
Immunosuppressive agents	Azothioprine[35]
Transplant agents	Mycophenolate[36-38]
Antimicrobials	Neomycin[39]
Chemotherapeutic agents	Busulphan
Vinca alkaloids	Colchicine, vincristine[40,41]
Antimetabolites	Methotrexate[42]
Angiotensin II receptor antagonists	Olmesartan[43]
Monoclonal antibody agents	Ipilimumab[44]

Usually, medication effects are completely reversible with cessation. Vincristine is a stathmokinetic agent causing mitotic blockade and prominent changes in the small intestinal mucosal crypts with prominent and readily visible metaphase arrest.

Table 3 shows an accumulated list of medication classes with examples of some currently used medications that cause sprue-like intestinal changes, potentially mistaken for celiac disease.[33] For each, removal of the offending agent (rather than institution of a gluten-free diet) results in clinical and histopathological improvement. In future, particular attention to emerging medications will be critical before attributing clinical and pathological changes to celiac disease.

EMERGING RISK ESTIMATES

Historically, studies initially suggested that celiac disease was detected mainly only in infancy and primarily in Europe or countries that experienced emigration (largely from the United Kingdom) to Canada and Australia.[45] Initially, it was believed that Ireland, in particular, had a high prevalence, particularly in western Ireland, specifically, Galway, up to 1 in 300 persons.[46] A similar experience had also been accumulated in some Scandinavian countries. In the United States, however, earlier reports suggested that detection of celiac disease was low.[45,47] In more recent years, however, these original perceptions have been altered dramatically. At least, in part, this development reflects widespread serologically-based case finding and screening, particularly in the United States.

Serologically-based studies have estimated that about 1% to 2% of populations in most countries evaluated have celiac disease, particularly in the United States and most European countries.[48-50] The precision of these studies is not clear, however, since standardization of serologically-based assays, including IgA tissue transglutaminase,[51,52] is limited. It has also been noted elsewhere that serological studies have likely overestimated sensitivity and underestimated specificity due to verification bias[53] as serologically-negative subjects are rarely biopsied.[54] However, these serological studies indicate that rates of undiagnosed disease are significant, even in Europe and United States. Prevalence data in these countries has recently been summarized.[54] In Sweden, children have rates of 1:285 and 1:77, Finland, 1:99 and 1:67, and Italy, 1:230 and 1:106. Similar rates have been noted in New Zealand, Australia, Argentina, and Israel.[55-61] In the United States, overall prevalence rates for children and adults were recorded at 1:104 and 1:105, respectively.[48,62] However, ethnic specific data are in the United States are limited. Hispanics appear to have a lower prevalence than non-Hispanics thought to be related to a low frequency of HLA-DR3, DQB1*0201 haplotype.[62] Similarly, celiac disease is rarely recorded in East Asian populations that lack this haplotype, but prevalence rates similar to Europe have been noted from the Middle East and South Asian populations. Interestingly, people of the Sahara in North Africa have the highest prevalence rate although Africans (and African Americans) have very low rates.[63] In many countries, there is limited or no data and it has been suggested that interpopulation differences in individual countries may not only reflect genetic factors, including HLA susceptibility alleles, but other environmental risk factors, including the geographic and temporal variation in nutritional practices.

Some of the most intriguing information has suggested a change in the prevalence of celiac disease in recent decades. Some believe that celiac disease may be increasing, particularly in North America and Europe. In part, some simply reflect increased physician recognition coupled with use of serologically-based testing for screening and case finding. However, a true change in prevalence may have occurred, possibly related to other confounding environmental variables.[11,64] In young male military recruits at Warren Air Force Base, a low prevalence was suggested based on evaluation of stored frozen sera collected from 1948 to 1952, compared to more recent control cohorts from Olmstead Country in 2006 to 2008.[64] In an independent report, increasing seroprevalence rates were also noted from 1974 to 1989.[65] Endoscopic biopsy, rather than serological screening has also been done. Routine duodenal biopsies obtained during endoscopic study defined moderate to severe architectural changes typical of celiac disease in 2% to 3% of adult Canadians referred for investigation.[66] Interestingly, in this study, environmental factors may have been important as a significant fall in new diagnoses of celiac disease occurred over 2 decades followed by a significant rise during the next decade.[66] Other long-term studies have also suggested a change in risk in recent decades[67,68] including a recent report from Hangzhou in China suggesting increased detection, possibly because of serological screening.[69] A case of celiac disease was also reported from Korea.[70] Similar biopsy-positive Asian Canadians with celiac disease were previously noted, including a Chinese woman.[71] A recent extensive study of HLA-haplotypes and wheat consumption in different regions of China also suggested that celiac disease occurs more frequently in China than currently reported.[72] A particularly high allele frequency of DQB1*0201 or DQB1*0201/02 occurs in Xinjiang in northwestern China, an area largely populated by Uygurs and Kazaks, rather than Han Chinese. Overall, calculated wheat consumption in China has also increased suggesting that the opportunity for gluten exposure is rapidly increasing. Interestingly, wheat consumption appears to be greater north of the Yangtze River along the ancient Silk Road compared to rice consumption regions south of the Yangtze River. Rural Xinjiang seems especially susceptible, as wheat consumption there is relatively high.[72] These significant and rapid changes in detection rates, defined by either increased or decreased rates based on use of either serologically-based methods or endoscopic biopsies would not likely reflect genetic factors, but instead, a response to other, possibly, superimposed environmental factors. Alterations in cereal production and processing, emergence of new or genetically altered forms of wheat or other grains, childhood infections associated with the so-called “hygiene hypothesis,”[73-75] breastfeeding or time after birth of initiation of feeding solids,[76,77] even changes in patterns of specialist referral and other factors, including medications, air pollution and cigarette consumption have all been considered. More studies are needed.

ALTERNATIVE AND NOVEL TREATMENTS

The gluten-free diet has been recommended for treatment of celiac disease for more than a half-century, as a result of the seminal early clinical studies by Dicke and colleagues from the Netherlands during and after World War 2, noted earlier.[18] However, gluten is ubiquitous and complete avoidance is difficult. In recent decades, per capita consumption of wheat and other processed foods that, in themselves, contain more gluten, have both increased. The gluten-free diet is costly, not universally available and compliance is difficult. A need for an alternative, or at least, added supplemental therapies that might reduce reliance on the gluten-free diet is evident.

A number of approaches have been considered (Table 4). Reduced exposure to gluten components in modern grains that are particularly immunogenic, for example, may be useful.[78] Modern hexaploid forms of wheat are believed to be more immunogenic, compared to ancient wild or diploid varieties of wheat.[79] Eventual development of genetically modified grains without significant numbers of immunogenic components may be possible, but this appears to be very challenging. Gluten contains many different immunogenic peptide sequences and some, but not all, of the genes responsible are not entirely known and located in different sites in the wheat genome.[80] Modification may result in a loss of the important baking features of the wheat and there remains a future potential for contamination with wild wheat strains.

Gluten could potentially be sequestered in the lumen with linear copolymeric binders effectively reducing exposure to the epithelium and limiting its effects. One copolymeric binder, hydroxyethylmethacrylate-co-styrene sulphonate, was shown to complex with gliadin reducing toxicity on intestinal epithelial

Table 4. Potential Alternative Forms of Therapy

Mechanism	Possible therapy
Reduced gluten exposure	Genetically-modified grains
	Copolymeric binders of gluten
Predigestion of gluten peptide	Proylendopeptidase (e.g., ALV003)
Tight junction blockade (zonulin)	Larazotide acetate (e.g., AT1001)
Transglutaminase 2 or HLA DQ2/DQ8 blockers	Development peptides
Immune tolerance induction	Peptide vaccination (NexVax)

cells *in vitro* and in a rodent model *in vivo*.[81,82] Human studies are still needed to determine if this approach has merit.

Another approach involves "pre-digestion" of dietary gluten. Ordinarily, ingested dietary proteins are hydrolyzed in the lumen by gastric pepsin and pancreatic proteases. In addition, enterocyte peptidases further hydrolyze resultant peptide products into amino acids, dipeptides and tripeptides for enterocyte transport into the portal venous system. Proline- and glutamine-containing peptides in gluten are resistant to enzyme proteolysis. As a result, only partial digestion of gluten occurs. It is thought that resulting peptides could induce an immune response in genetically-programmed individuals leading to celiac disease.[83,84] Prolylendopeptidases derived from some plants and different bacteria (i.e., *Flavobacterium*, *Sphingomonas*) can hydrolyze internal proline-glutamine bonds in a proline-containing peptide[85] but may be subsequently inactivated in the acidic milieu of the stomach.

Prior studies using a combination of a barley-derived endoprotease and prolyl-endopeptidase in powder or tablet form appeared to be stable and caused breakdown of wheat gluten with reduced immune effects.[86-88] Prolylendopeptidase activities derived from *Aspergillus niger* were shown to inhibit a gliadin-stimulated immune response by gluten-specific T-cells.[89] In a model system, the majority of hydrolytic activity occurred in the gastric compartment with only limited activity needed in the small intestine.[90]

Specific enzymes derived from other microbial species, have also been shown to be operational in a gastric environment, and have been cloned and characterized.[91-94] As shown in Table 5, clinical trials were done using ALV003, a novel combination glutenase recombinant orally-administered product. Two of these trials (NCT00959114 and NCT01255696) were published as a

Table 5. Treatment Trials for Celiac Disease

Therapy	NCT ID	Sponsor	Status*
ALV003	01255696	Alvine	Complete
AN-PEP	00810654	VU Med Ctr	Complete
AT1001	01396213	Alba	Complete
ALV003	00959114	Alvine	Complete
AT1001	00620451	Alba	Complete
AT1001	00492960	Alba	Complete
AT1001	00889473	Alba	Complete
AT1001	00386165	Alba	Complete
ALV003	00859391	Alvine	Complete
AT1001	00362856	Alba	Complete
ALV003	00626184	Alvine	Complete
NexVax	008879749	Nexpep Pty	Complete

ALV003 and AN-PEP, prolylendopeptidases; AT1001, larazotide acetate; NexVax, vaccine for immune tolerance induction.
*Listed as completed on clinicaltrials.gov.

randomized, double-blind, placebo-controlled phase 2 trial[95] demonstrating that ALV003 attenuates gluten-induced small intestinal injury in patients with celiac disease in the context of a "gluten-free" diet daily containing up to 2 g gluten (equivalent to approximately one-half standard slice of bread in the United States).

A different variation on this "enzymatic" approach was evaluated in a pilot study of gluten-free sourdough wheat baked goods employing lactobacilli and fungal proteases causing a gluten content of <8 ppm and seemed safe for young celiacs with no changes in hematological, serological or intestinal permeability end points.[96] Another study compared natural flour baked goods to two hydrolyzed baked good groups. Most in the latter two hydrolyzed groups had no clinical, serological, or histological worsening.[97] Others have employed probiotic preparations, specifically VSL#3, a mixture of lactic acid and bifidobacteria in preliminary studies showing effective hydrolysis of gliadin peptides implicated in celiac disease[98] combined with evidence of increased barrier function associated with enhancement of tight junction markers in an animal model.[99]

A further therapeutic approach has focused on prevention of epithelial tight junction passage of molecules, specifically immunologically-active gluten peptides. In celiac disease, including its most early phases, it has been hypothesized that the intestinal mucosa is "leaky" with increased paracellular permeability. Zonulin is a specific tight junction protein highly expressed in celiac disease that functions together with other transmembrane proteins to regulate permeability of the epithelial barrier.[100] Gliadin is thought to bind to the chemokine receptor CXCR3 releasing zonulin and leading to increased intestinal permeability.[101] Larazotide acetate (AT1001) is a peptide that antagonizes zonulin by receptor blockade[102] and is believed to impair paracellular transport from gliadin peptides and their resultant immunological effects. To date, clinical trials have suggested that larazotide appears to be safe with some symptomatic benefit compared to placebo, but no significant change in intestinal permeability.[103] As shown in Table 4, the phase 2 clinical trial has been completed (NCT01396213).

Once gluten peptides pass through tight junctions, tissue transglutaminase 2 enzyme-induced deamidation occurs. The deamidated peptides each assume negative charges causing an enhanced affinity for the binding grooves on HLA-DQ2 and/or DQ8 molecules located on antigen-presenting cell surfaces. As a result of this process, T-lymphocyte activation and subsequent histopathologic mucosal effects occur. To counteract these effects, different hypothetical approaches have been considered.

One approach may involve blockade of transglutaminase 2 or the specific HLA-DQ2 and/or DQ8 molecules to prevent this peptide binding and resultant immune-related mucosal inflammatory effects. Inhibition of *in vitro* transglutaminase 2 activity inhibits gliadin-specific T-cell clones from patients with celiac disease as well as gliadin-induced proliferations of some types of mucosal lymphocytes.[104,105] Development of gluten peptide analogues may hypothetically act as HLA-DQ2 blocking agents by prohibiting binding or access to the binding grooves on antigen presenting cells is another area being considered.[106]

Another novel approach is immune tolerance induction. A peptide vaccine that could promote tolerance of some immunologically-active mucosal cells involved in the pathogenesis of celiac disease may be possible. Nexvax peptide vaccine employs three different gluten peptides to can hypothetically lead to tolerance in celiacs. Prior studies in HLA-DQ2 transgenic mice with gluten-sensitive T-cells demonstrated efficacy while patients with celiac disease treated with this agent demonstrated acceptable safety and anti-gluten T-cells. Further studies to evaluate efficacy and long-term safety in humans with celiac disease for all of these therapeutic options are clearly needed.

CONFLICTS OF INTEREST

No potential conflict of interest relevant to this article was reported.

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