REVIEW ARTICLE

Veracity and Fidelity of Cone Beam Computed Tomography while Evaluating the Thickness of Gingiva- A Review

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ABSTRACT

The purpose of this review is to evaluate the competence of cone-beam computed tomography in estimating the thickness of gingiva. PubMed database was searched for studies conducted in the period from 1st January 2005 to 30th June 2020 for the procurement of the potentially relevant abstracts. The search was conducted by two methods; the first one includes only Medical Subject Headings (MeSH) and the second one includes the keywords which have a similar meaning. Appropriate articles which we're highlighting the aim of the study and fulfilled the inclusion criteria were included in this study. 343 articles were obtained while searching the PubMed database, out of which 50 articles were included on a title basis. After that, on the abstract basis, only 19 articles were extracted and finally, only 12 articles were fulfilling the inclusion criteria. Cone-beam computed tomography (CBCT) is a simple and non-invasive method, which shows guaranteed results in the measurement of the thickness of the gingiva. Nowadays, CBCT is considered an authentic and more appropriate technique used by clinicians for determining the association between the various structures supporting the periodontium.

Keywords: CBCT, Gingival thickness, Soft Tissue CBCT, Noninvasive technique, Cone-beam computed tomography, Gingival Biotype.

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INTRODUCTION

The discrepancy in periodontal anatomy has an association between the alveolar bone and surrounding tooth structures as well as a probable association to periodontal health and disease [1,2]. Ochsenbein and Ross [3] constituted that there were two types of gingival contour which follow the underlying bony contour and are related to the shape of teeth, one is associated with triangular and tapered teeth, with festooned margin is "thin scalloped gingiva" and another related with squarish shape of teeth and flattened marginal gingiva is "thick flat gingiva". In 1989 Seibert and Lindhe [4] coined the term "Periodontal Biotypes" and they also elucidate the discrepancy concerning the shape and length of teeth along with morphology of gingival tissue and underlying bone. While compiling the literature, morphology of gingiva could be classified into three significant groups: (a) triangular and tapered teeth, the contact point is more towards incisal, slight cervical protuberance, scanty keratinized tissue, thin gingiva followed by thin underlying bone all these belong to a thin scalloped type. (b) the squarish shape of teeth marked cervical protuberance, the contact point is shifted apically, ample keratinized tissue, thick gingiva followed by thick underlying bone belongs to thick flat. (c) teeth are slender, the gingiva is thick, highly scalloped gingiva, scanty keratinized tissue belongs to thick scalloped [3-7]. Periodontal biotype appears to be an important feature while contemplating the inference of the various dental procedures especially in the treatment of periodontal disease. Inflamed periodontium severely affects the clinical conditions it leads to the increased depth of pocket in case of thick flat gingival biotype and leads to recession in case of thin scalloped gingival biotype [8].

Visual Examination is one of the simple and oldest method for examining the gingival biotype. The effectiveness of this method is evaluated by the clinician's experience [9]. Thick gingival biotypes were easily recognized as compared to thin ones. Another method used for measuring gingival thickness is the ultrasonic device propound by Müller in 2007 [10]. Inference indicated that the Ultrasonic Device method is more authenticated but not reliable in identifying small variations [11]. Another more effective method for evaluating the gingival thickness is Transgingival Probing[8] in which an endodontic file or periodontal probe is used. This method is more precise and was used to assess the outcome of guided tissue regeneration [12]. Gingival thickness was assessed by sounding the facial facet of bone with the help of a periodontal probe [13]. If the probe is visible while bone sounding then the biotype is thin if not then the gingival biotype is thick [14]. Three-dimensional radiographic techniques, such as cone-beam computed tomography (CBCT) have been introduced for the image analyses of the maxillofacial region [15]. CBCT technology offers high-quality diagnostic images for the clinician and has become an essential tool in dentistry [16]. Soft tissue CBCT is a pioneering technique that is derived from CBCT to envision and actual evaluation of the distance between the hard and soft tissue. This is a non-invasive and simple method, with the help of this method clinicians will be able to determine the association between the various structures supporting periodontium [17]. The purpose of this review is to evaluate the competence of cone-beam computed tomography in the estimation of the thickness of gingiva.

MATERIAL AND METHODS

Search strategy
PubMed database was searched for studies conducted in the period from 1st January 2005 to 30th June 2020 to find potentially relevant articles for this review. The search was conducted by two methods; the first one includes only Medical Subject Headings (MeSH) and the second one includes the keywords which have a similar meaning. In the first method we searched for the "Cone Beam Computed Tomography" and "Gingival Thickness". and in the second method, we searched these keywords "cone beam computed tomography" or "CBCT" or "digital volume tomography" and "Gingival thickness" or "Gingival biotype" and then results procured from the first and second methods were combined and common ones were taken only once. (Table 1)

Eligibility criteria
The search for the articles was limited to the following inclusion criteria: 1) should be original articles, not a review one. 2) only clinical studies which were conducted in the human population were included. 3) It should examine the

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accuracy of Cone Beam Computed Tomography in measuring the gingival thickness. 4) only articles written in the English language were considered.

Screening and selection

Appropriate articles were screened firstly based on titles; irrelevant titles were excluded from the study, then selected articles were screened based on abstract by keeping in view of inclusion criteria. Then full-text of selected articles which were fulfilling the inclusion criteria were procured in this study. All the articles were screened independently by two reviewers, in case of discrepancy third reviewer's judgment was considered final. For assessing the risk of bias both at the study level and at the outcome level, we used a checklist for assessing a diagnostic or predictive test developed by Leake [18] after modification. This was performed by two reviewers independently; finally, disagreements were resolved by discussion between the two reviewers; if no agreement could be reached, it was planned that a third author would decide. The following were the questions included in the checklist:

1. Was there a clear question for the study to address?
2. Were the exclusion and inclusion criteria mentioned?
3. Was the sample size sufficient?
4. Was the test clearly described?
5. Did the test report the technical parameters that may influence the results?
6. Was the test evaluated in a valid clinical setting?

RESULTS

The results were summarized in a flow diagram (figure 1). A total of 343 articles were procured while searching the PubMed database, out of which 50 articles were included at the title stage and 293 articles were excluded. After that, at the abstract stage, only 19 articles were extracted that were relevant and related to our main aim and 31 articles were excluded. Only 12 articles were fulfilling the inclusion criteria which were included for this review; they were published between 1st January 2005 to 30th June 2020. Meanwhile, 7 articles were also excluded out of which 4 were written in other languages and 3 of them were animal studies and some studies on cadavers. The data were extracted from the 12 included studies were made (Table 2). Then, these studies were scored using the previously mentioned checklist (Table 3). Finally, the results of the criticism revealed that no studies scored <50%, while when it comes to the meta-analysis, all should be included in the results; however, in the end, we found that no meta-analysis for the collected data could be made due to the lack of complete and detailed statistical presentation of the CBCT measurements for gingival thickness.

On analyzing the data pooled from the studies included in this review, we have seen that Borges et al.[21]assessed CBCT asa potent diagnostic method to envision and evaluate gingival thickness and distance between gingival margins and vestibular and interproximal bone crests. Two studies have used CBCT in assessing palatal mucosa, which was conducted by Ueno et al.[20]and Yilmaz et al.,[22]both analyzed the palatal mucosal thickness and used CBCT for the identification of the greater palatine foramen location and palatine groove. A study conducted by Mallikarjun et al.[24]in which they compared cone beam computed tomography and RVG for the gingival thickness in the anterior maxilla. But no statistically significant differences were achieved while comparing the above. They concluded that measurement of hard and soft tissue cone beam computed tomography and RVG methods can be used. In another study conducted by Silva et al.[26] in which they assessed gingival tissue and bone on the labial surface of the anterior maxilla and compared CBCT values with clinical probing. They find CBCT as a noninvasive technique for the measurement of gingival thickness. Alves et al.[27] found Computed Tomography reliable and clinically useful for assessing the biotype of gingiva via computed tomography scanning (CTS) along with the transparency of periodontal probe, transgingival photography.

DISCUSSION

While trying to find an answer to our question, we found that the CBCT is effective in measuring gingival thickness. However, in the included studies, we tried to highlight the proficiency of the noninvasive method CBCT to envision, assess the dimensions, and evaluate the association of several structures of the periodontium and dentogingival attachment apparatus while comparing it with other invasive and non-invasive methods like visual evaluation, transgingival probing, ultrasonic device and radiovisiography (RVG). Visual inspection is one of the possible methods of inspecting gingival thickness but this method is not reliable and also requires expertise and experience. This method is also not reliable in the esthetic areas. One of the old and most reliable method of measuring gingival thickness is transgingival probing, no specific equipment is required. This method is invasive as it requires local anesthesia and is quite uncomfortable for the patient.[30-32] This method has certain limitations as it can be done while using a rubber stopper on the endodontic file or probe for bone sounding and which might slip while performing this, which misleads the measurement.

The ultrasonic device has been suggested as a more effective method than transgingival probing. Eger et. al.[11] concluded that an ultrasonic device is more reliable and valid in measuring the gingival thickness with a resolution of 0.1mm. There is an issue about transducer probe diameter due to which it is quite difficult to use it in the posterior region. Besides CBCT is widely used in assessing hard tissue, due to its high diagnostic accuracy used in assessing soft tissue. Januario et al.[17] demonstrate that gingival thickness was measured by using the ST-CBCT technique in which CBCT scans were taken by retracting lip and tongue. In another study conducted by Yilmaz et al.,[22] the palatal thickness was measured and high contrast and resolution medical LCD displays were used to detect the soft tissue structures to make a proper measurement while using CBCT. It is important to mention that this is a quantitative assessment method and not a qualitative assessment because differentiation of particular macro and microscopic characteristics of the tissues cannot envision such as inflamed gingiva would have a similar appearance on the Soft Tissue CBCT scans as healthy gingiva. In a similar manner, it is not possible to differentiate between gingival connective tissue and gingival epithelium which shows a similar appearance on the Soft Tissue CBCT scans [17].

CONCLUSION

It can be conducted that CBCT is a noninvasive method that has the most promising results regarding the measurement of gingival thickness. CBCT is considered a reliable and more convenient technique for clinical diagnosis of gingival phenotype which enables the clinician to avoid complications in Periodontology, Implant Dentistry in the esthetic zone, Prosthetic Dentistry, Operative Dentistry, and Orthodontics.

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Table 1: The search strategy developed using the PubMed database

Search Method	Terms/Query	Item found
1	Cone beam computed tomography (MeSH*)	14076
2	Gingival thickness (MeSH*)	1148
3	Cone beam computed tomography and Gingival thickness	85
4	Cone beam computed tomography or CBCT or digital volume tomography	16599
5	Gingival thickness or Gingival biotype	1255
6	Cone Beam Computed Tomography or CBCT or digital volume tomography and Gingival thickness or Gingival biotype	343

*MeSH – Medical Subject Headings

Table 2: Summary of the findings of the articles included in the study

Author	Title	Findings
Januário et al. (2008)[17]	To establish ST-CBCT* to enhance the image of soft tissue and find the association of the structures of dentogingival unit and also measures dimensions.	ST-CBCT scans showed a clear presentation of the soft tissue, easy to measure the dimensions, and relationship of the structures of the periodontium and dentogingival attachment apparatus.
Cooket al. (2011) [19]	To examine the differences in the thickness of the labial plate in patients with thin and thick biotypes.	Periodontal biotype is significantly associated with labial plate thickness, keratinized tissue width, alveolar crest position, probe visibility, and gingival architecture but no association with facial recession.
Ueno et al. (2014) [20]	To evaluate the thickness of palatal mucosa from the margin of the gingiva to the mid-palatine suture in the Japanese population while using CBCT**.	Palatal mucosa was thickest at 9 to 12 mm from the gingival margin in canine to premolar regions. CBCT** has been used to identify greater palatine foramen and palatine groove which can help in diagnosing the palate to diminish surgical complications.
Borgeset al. (2015) [21]	To evaluate CBCT** as a diagnostic method to determine the thickness of gingiva and distance between gingival margin and vestibular and interproximal bone crests.	CBCT** is a potent diagnostic technique to envision and evaluate gingival thickness and distance between gingival margin and vestibular and interproximal bone crests.
Yilmazet al. (2015) [22]	To assess the thickness of palatal mucosa and location of greater palatine foramen using Cone-beam computerized tomography.	Palatal mucosal thickness from 2nd molar to canine were 3.7, 3.3, 3.7, 3, and 3 mm, respectively. There was no association between the palatal junction angle and the palatal depth. Greater palatine foramen was observed at the level of a third molar tooth, between third and second molar, second molar.

Maria et al. (2016) [23]	To explore the association between periodontal morphometric parameters and to develop the gingival biotype based on the variables by using CBCT**.	The thickness of gingiva at CEJ was positively associated with the thickness of labial bone plate and crown form. The crown form was not associated with the thickness of labial bone plate.
Mallikarjunet al. (2016) [24]	To evaluate the gingival thickness of the maxillary anterior and its association with underlying alveolar bone thickness by comparing radiovisiography and CBCT**.	While comparing radiovisiography (RVG) and CBCT** no statistically significant differences were procured. They also failed to disclose any significant association between the gingival width and the alveolar bone width in the maxillary anterior region.
Esfahanizadeh et al. (2016) [25]	To determine the thickness of soft tissue and buccal bone and their association in the maxillary anterior region using CBCT**.	The mean buccal bone and soft tissue thickness in the anterior maxilla was <1mm and there was a mild linear correlation between them.
Silva et al. (2017) [26]	To assess the influence of lip retraction on CBCT** measurements of bone and gingival tissues on the labial surface of the anterior maxilla.	Gingival tissue thickness obtained from CBCT** scans with lip retraction showed significant correlations with those obtained clinically.
Alveset al. (2018) [27]	To evaluate the validity of gingival biotype using CTS***.	The comparison between CTS*** and the transgingival method shows high specificity and low sensitivity for defining thin biotypes.
Shao et al. (2018) [28]	To examine the distribution of periodontal biotype in the young Chinese population and also measure the gingiva thickness by different methods.	Thick-flap type with low aesthetic risk is the most common periodontal biotype in the young Chinese population.
Gürleket al. (2018)[29]	To assess the thickness of Soft tissue by using cone-beam computed tomography and to co-relate with the ultrasonic device and transgingival probing measurements.	The thickness of soft tissue depends on the measuring method and location. The high agreement between CBCT and transgingival probing has promising results for measurements of soft tissue thickness.

*ST-CBCT- Soft Tissue Cone-Beam Computed Tomography
**CBCT - Cone beam computed tomography
***CTS-Computed tomography scanning

Table 3: Checklist scoring

Name of authors and year	Checklist scoring
Januário et. al. 2008	5/6
Cook et. al. 2011	4/6
Ueno et. al. 2014	5/6
Borgeset. al. 2015	5/6
Yilmaz et. al. 2015	4/6
Maria et. al. 2016	5/6
Mallikarjunet. al. 2016	3/6
Esfahanizadeh et. al. 2016	5/6
Silva et. al. 2017	5/6
Alveset. al. 2018	5/6
Shao et. al. 2018	3/6
Gürlek et. al. 2018	4/6

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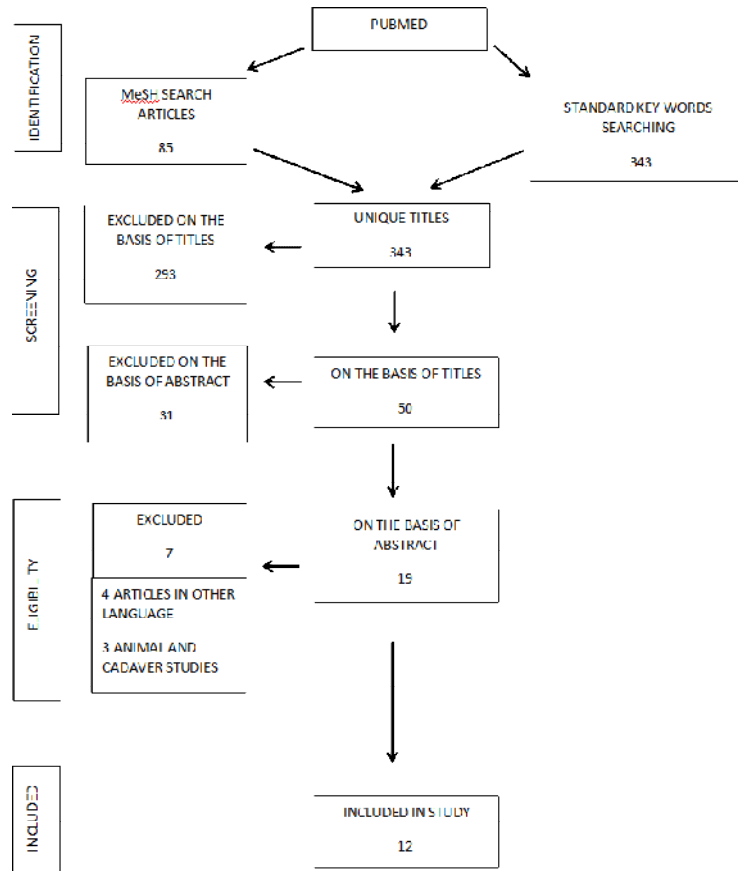
Figure 1 Flow diagram summarizing results

RESEARCH ARTICLE

Utilization of Fish and Shellfish Waste as Ingredients for Aquaculture Feed

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ABSTRACT

In recent years, fish waste management has become a global issue. Disposal of seafood waste pollutes the environment. Waste from seafood includes small fish, head, viscera, fins skins which are not suitable for human consumption. The seafood waste can be formulated to produce feed for animal and fish as a supplement. These wastes are a rich source of protein, minerals and vitamins which can overcome nutrient deficit in the animals when added to their diet. Fish silage, fish meal and fish oil are by-products prepared from fish waste. Fish oil in particular provides the triglycerides of fatty acids and phospholipids in animal feed, whereas protein hydrolysates supply a significant amount of nitrogen. These are also utilised in the production of pet food and fertilisers. Fermentation, biotechnology, and bio preservation processes are used to prepare these by-products for feeding.

Keywords: Fish processing waste, Fish meal, Fish silage.

INTRODUCTION

The development of new creative products from fish processing waste benefits the environment by reducing the problem of waste disposal, it also increases revenues of the processor. The biggest advantage is nutritional, as fish waste accounts for half of the industry's raw material volume and is a low-cost source of nutrients (Oetterer, 2002).Transporting fish waste to by-products processing factories is not always a sustainable operation(Nunes, 1999). When it comes to energy conservation, companies must develop techniques for utilising wasteas it causes major problem of environment pollution. The ideal solution is maximum utilization of the raw materials and production of a by-product from waste (Maia et al., 1998). The waste-to-energy conversion technologies have not been adopted commercially due to its higher initial cost.

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The unpleasant odour makes it impossible for sanitary landings and effluent treatment ponds as an alternative in areas of interior and coastal waterways, as these water are mostly used for recreational purpose. Despite all the efforts and campaign been made on the nutritional properties and consumption of fish, there will always be some portion which are regarded as insufficient due to personal choices, sizes, appearance and exceed the processing capacity (Lustosa Neto, 1994). The transformation of fish waste into a product that may be used as an element in animal diets is one technique to reduce the environmental difficulties caused by the excessive amount of fish waste (Ristic et al., 2002). The goal of this review was to discuss about how to use fish waste, how to make chemical silage, and how to use it as an aquaculture feed ingredient.

Uses of marine processing waste

The manufacturing of animal feed from waste seafood products is important for minimising pollution and offering low-cost animal production (Westendorf et al., 1998; Westendorf, 2000).The processing wastes are easily available as they are not fit for human consumption and due to the problem of disposal. When converted these waste becomes a rich source of protein of low cost feed ingredients for aquaculture, poultry and cattle (Esteban et al., 2006). Lipids, minerals, proteins, and fats are found in the waste of mackerel, croaker, flying fish, and sardine (Rustad, 2003; Khan et al., 2003).

Fish meal

Fish meal is made by drying whole fish or undesirable fish parts and grinding them into a powder. Menhaden, capelin, and anchovy are the most commonly utilised fish for fish meal (Hevroy et al., 2004). This fish meal is combined with other ingredients to make fish and shrimp feed.In fish meal, the main nutrient is protein constitute about 70%, 10% of minerals, fats constitute 9%, water content is 8% and the remaining consists of vitamin, ashes, pantothenic acid and a variety of other minerals. It can be divided into several quality levels based on freshness, solubility, amino acid types, and processing method (Barlow and Windsor, 1984; Babbitt, 1990; Gildberg, 2002). Advancements in fish meal production processes have resulted in the development of new processing equipment which requires lesser time as compared to the conventional methods. Fish meal is the main component incorporated in aquaculture feeds, cattle, pig, ruminants, pets and poultry.Kriton et al., 2018 investigated whether total or high substitution of sustainable plant raw materials (plant meal and oils) in long-term feeding for rainbow trout, gilthead sea bream, and common carp can cause deterioration during ice storage.

Fish oil

Fish oil, which can be derived from discarded fish and unwanted fish products, is also a major component of fish and crustacean feed. The amount is usually determined by the fat content of the specific fish species. Fish normally has a fat content of 2-30%. Nearly half of the body weight produced as waste during the processing of fish will be a significant source of highquality fish oil for human consumption or biodiesel generation. Fish oil contains the main fatty acids docosahexaenoic acid (DHA) and eicosapentaenoic acid (EPA). They are polyunsaturated fatty acids known as omega-3 fatty acids. They're mainly found in marine species that have a lot of polyunsaturated fatty acids (Zuta et al. 2003).

Fish bones

The backbone and cartilage of fish contain a lot of calcium phosphate, minerals, and about 30% protein. To obtain protein hydrolysates, scientists used a variety of enzymes, including proteases, chymotrypsin, and neutrases, on the backbone of a tuna fish (Je et al., 2007). These bone Protein hydrolysates are effective antioxidant agents and can be used to make feed (Morimura et al., 2002). Many natural antioxidants and germicides are now employed to improve food quality, and many are derived from food waste (Ucak et al., 2018). Seaweeds contain a variety of polyphenols, flavanols, and flavanol glycosides. When these substances are included in diet, they can have a positive impact on human health (Topuz et al., 2016). Unwanted fish products are sorted into dry, liquid, fresh, and frozen forms, which are then used to benefit agriculture. After composting these small fish with huge amounts of carbon sources such as sea shells and fish bones, these products are used as a fertiliser in agriculture. These undesired substances are also utilised in fish and crustacean feed and as a fish trap.

Fish silage

Fish silage is a liquid product made from the whole or sections of a fish, with acids, enzymes, or lactic acid producing bacteria added, and the bulk liquefied by the action of the fish's enzymes (FAO, 2003). A. I Virtanen invented acid silage in 1920 to preserve forages by utilising hydrochloric and sulphuric acid. In 1936, hydrochloric, sulfuric, and formic acids, as well as sugars, were used in fish experiments in Sweden (Tatterson and Windsor, 1974). Organic acids, like formic acid, are more expansive than mineral acids in general. Organic acids, like formic acid, are more expansive than mineral acids in general. They do, however, produce fewer acid silage that do not require neutralisation prior to use. It is necessary to consider bactericide action. It has been suggested that a mixture of formic and propionic acids be used. Preservation reagents for chemical silage are inorganic acid, organic acids, or a mixture of both organic and inorganic acids, which are slightly more acidic than formic acid but do not require neutralisation before use. Because of their inexpensive cost, inorganic acids such as hydrochloric acid and sulphuric acid are recommended (Oetterer, 2002).

Use of fish silage

Fish meal can be replaced with fish silage. In comparison to fish meal, silage production has the following advantages: the technology is simple, scale-independent, can be used on-site, low investment, the process reduce effluents. The products needs to be dried as consumption in pastry form is voluminous (Kompiang, 1981; Beerli et al., 2004). A lot of attention has been paid to the use of fish silage in fish feeding. Because of its similarity to the raw material and low cost, silage has a lot of potential in aquaculture (Goddard and Perret, 2005; Hussain and Offer, 1987; Fagbenro et al., 1994; Vidotti et al., 2003). Microbial fish silage when added with soumeal, bone meat powder and poultry by-products into diet of *Oreochromis niloticus* and *Clariusgariepinus,* there is no significant difference in growth performance, carcass composition and protein consumption with diets prepared using fish meal as a source of protein (Fagbenro and Jauncey 1988; Fagbenro et al. (1994).

Uses of by-products of shellfish

Shrimp waste meal, which includes the includes the heads, appendages, and exoskeleton, is high in lysine. Shrimp meal can be in place of fish meal in the composition of broiler feed. The presence of lysine and methionine in shrimp waste makes it protein higher quality as compared to fish meal (Fanimo et al., 2000). Squid contain high quality protein (Gokoglu et al., 2017). Oyster shells are a great source of protein for animal feeds (Sathiadhas and Aswathy, 2004). Red crab are not consumed by human beings due to their smaller sizes but are a great additives to animal feed for their highnutritional content. It provides a significant amount of protein and minerals (Villarreal et al., 2004), who found favourable results for the American lobster when utilising feed supplemented with red crab meal. Red crab is also high in -carotene and two astaxanthin esters (Wilkie, 1972).

Chitin

Exoskeletons of crustaceans include 15-20 percent chitin by dry weight, making it a structural component. The synthesis of chitin and chitosan from food waste (crustacean canning) has proven to be both environmentally and economically viable, particularly when carotenoids are recovered. The wastes contain significant levels of chitin, which is marked as a fish food additive (Arvanitoyannis, 1999; Kumar, 2000). Coward-kelly et al. (2006) treated shrimp head waste (*Penaeus indicus*) with lime at different temperatures (75, 100 and 125°C) and lime/shrimp ratios (0, 0.05, 0.1, 0.2 g $Ca(OH)_2$ per gram dry shrimp to determine the repeatability, effect of temperature, and effect of lime loading on solubilizing protein in shrimp head waste. Shrimp heads were hydrolyzed in less than 15 minutes and not need any special treatment (low temperature, low lime loading and short times). 20 % ash, 10.3% total Kjeldhal nitrogen (64% crude protein and chitin), 18 % lipids and other compounds, and little amino acid degradation were found in shrimp head waste. The protein rich material can be utilised as a monogastric animal feed supplement, with the residual solid – which is high in calcium carbonate and chitin- being used to manufacture chitin and chitosan. The latter can be used as a dietic product or as edible films for food preservation in a variety of ways, either alone or in combination (Arvanitoyannis et al., 1997, 1998).

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Environmental and economic impact

Fish waste management has become a concern that has a negative impact on the environment (Arvanitoyannis and Kassaveti, 2008). As a result, Fish waste management has become a concern that has a negative impact on the environment (Arvanitoyannis and Kassaveti, 2008). As a result, those methods are utilised for waste management that are both inexpensive and environmentally friendly. Fish meal, fish oil, fish silage and organic fertilisers are examples of these strategies.

CONCLUSIONS

The aim of this review is to describe how fish processing waste can be used in animal feed and aquaculture feeding. Various biotechnology and biopreservation approaches can be utilised to extend the life of seafood wastes. The intake of wastes in this manner compensates for the protein and other nutrients deficient in the animal feed industry's processing. The conversion of waste material and subsequent reutilization can provide economic benefits to the sector while also addressing a major issue with waste discharge, which is a polluting material. The usage of waste in the fish industry, as well as the cost of rations, will be major economic factors (Borghesi, 2004; Ferraz de Arruda et al., 2006).

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RESEARCH ARTICLE

A Study on Pythagorean Fuzzy Graph in Traffic Flows

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ABSTRACT

In this paper, the new idea of finding the causes of accidental zone in traffic flow using vertex colouring of Pythagorean fuzzy graph and examined the chromatic number of a graph with an illustrative example.

Keywords: Pythagorean fuzzy graph, Vertex coloring, Traffic flow, Accidental zone, Chromatic number.

INTRODUCTION

Yager (2013) introduced a Pythagorean fuzzy set (PFS), whose origin from great philosophers, mathematicians named Pythagoras. Yager suggests that PFS is useful for decision making purpose. Garg (2018) explored an idea of Pythagorean fuzzy sets in decision-making. Later he proposed concept of Pythagorean fuzzy set (PFS) has been extended to interval-valued Pythagorean fuzzy set and hesitant Pythagorean fuzzy set. Graph theory plays a vital role in the area of mathematics and has numerous applications in day-to-day life such as in area of modern science and technology, transportation system, data mining, image capturing, social network. The graph theory was first introduced by Swiss mathematician Leonhard Euler (1735) by solving a Konigsberg bridge problem. Zadeh (1965) introduced fuzzy set which has been implementing in various fields like group decision, engineering etc. Fuzzy graph was found by Rosenfeld (1975), he described that fuzzy analogues of different basic graph-theoretic ideas like bridges, paths, cycles, trees, connectedness and some properties.

Krassimir T. Atanassov (1998) was established the intuitionistic fuzzy set (IFS) which is generalization of the fuzzy set. Later he extended various concepts and properties of intuitionistic fuzzy set (IFS). Krassimir T. Atanassov (1999), were introduced the concept intuitionistic fuzzy graph. Parvathi.R and Karunambigai.M.G (2006) gives a new definition to the intuitionistic fuzzy graph and describe its properties. Muhammad Akram et al. (2019) extended the definition of Pythagorean fuzzy sets (PFS) to Pythagorean fuzzy graphs (PFG) and its properties, and studied the regularity of Pythagorean fuzzy graph (PFG) product. The Pythagorean fuzzy graph is an extension of intuitionistic fuzzy graph and it provides an accurate value for the problem which is vague. Francis Guthrie's (1852) four color

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conjecture introduced a graph colouring in graph theory. Graph coloring has wide application in the area of telecommunication, networking, scheduling, timeline, etc.

Susana Munoz et al. (2004) proposed two approaches to graph coloring in fuzzy graph. The first approach is to generalize the concept of chromatic number of graph. The second approach is based on an extension of coloring function by defining distance between colors of timetabling problem is analysed using exact algorithm for obtain chromatic number. Myna.R (2015) represents the traffic network of city using application of fuzzy graph in traffic. Muhammad Akram et.al. (2019) extended the concept of Pythagorean fuzzy graph from Pythagorean fuzzy set, Pythagorean fuzzy graph help us to find the solution for vague concept. Yamuna et al. (2020) developed the concept of Pythagorean Fuzzy Graph Vertex Coloring with an illustrative example, Pythagorean fuzzy graph act as a tool in the area of decision making. In this paper, we discussed about one of the applications of graph theory in real life (i.e.,Traffic flow in a city) by using Pythagorean fuzzy graph and we have discussed various accidental zones in a city with an illustrative example.

Preliminaries

Definition 2.1. A ***Fuzzy graph*** $G = (\sigma, \mu)$ is a pair of functions $\sigma : V \rightarrow [0,1]$ and $\mu : V \times V \rightarrow [0,1]$, where for all $u, v \in V$, we have $\mu(u, v) \leq \sigma(u) \wedge \sigma(v)$.

Definition 2.2. An ***Intuitionistic fuzzy graph*** (IFG) is of form G = (V, E) where

1. $V = \{v_1, v_2 ... v_n\}$ such that $\mu_1 : v \rightarrow [0,1]$ and $\gamma_1 : v \rightarrow [0,1]$ denotes the degree of membership and non-membership of the element $v_i \in V$ respectively and $0 \leq \mu_1(v_i) + \gamma_1(v_i) \leq 1$ for every $v_i \in V$, (i = 1,2,..n)

2. $E \subset V \times V$ Where $\mu_2 : V \times V \rightarrow [0,1]$ and $\gamma_2 : V \times V \rightarrow [0,1]$ such that
$$\mu_2(v_i, v_j) \leq \min [\mu_1(v_i), \mu_1(v_j)]$$
$$\gamma_2(v_i, v_j) \leq \max [\gamma_1(v_i), \gamma_1(v_j)]$$
And $0 \leq \mu_2(v_i, v_j) + \gamma_2(v_i, v_j) \leq 1$ for every $(v_i, v_j) \in E$.

Definition 2.3. A ***Pythagorean fuzzy graph (PFG)*** on a non-empty set S is a pair $G = (M, N)$ with M a Pythagorean fuzzy set on S and N a Pythagorean fuzzy graph on S such that $\tau_N(xy) \leq \tau_M(x) \wedge \tau M(y), \varphi_N(xy) \geq \varphi_M(x) \vee \varphi_M(y)$ *and* $0 \leq \tau^2{}_N(xy) + \varphi^2{}_N(xy) \leq 1$ and for all $xy \in S \times S$, where the membership function of N is $\tau_Q : S \times S \rightarrow [0,1], \varphi_Q : S \times S \rightarrow [0,1]$ is the non-membership function of N.

Definition 2.4. A ***k-vertex coloring*** of a graph G is an assignment of k colors, 1,2,...,k, to the vertices of G. The coloring is proper if no two distinct adjacent vertices have the same color. The ***chromatic number*** $\chi(G)$ of a graph G is the minimum k for which, G is k-colorable.

Definition 2.5. The ***Pythagorean fuzzy graph*** $G(\sigma, \gamma)$ consisting of a family $\psi = \{\psi_1, \psi_2, ..., \psi_k\}$ of Pythagorean fuzzy set on a set P is called a K-vertex coloring if

(i) $\max \psi_i(x) = \sigma, \forall x \in \sigma$

(ii) $\min(\psi_i, \psi_j) = 0$

(iii) $\min \{\psi_i(\alpha_i(x)), \psi_i(\alpha_i(y))\} = 0$ and $\max \{\psi_i(\beta_i(x)), \psi_i(\beta_i(y))\} = 1, (1 \leq i \leq k), \forall$ strong edge of G.

The minimum colors for G with k-vertex color is called the coloring number of the Pythagorean fuzzy graph G, or the Pythagorean fuzzy coloring number and it is denoted by $\chi(G)$.

Expressing the traffic flow problem using pythagorean fuzzy graph:
Graph theory has a wide application in various disciplines. A graph is a pictorial way of representing two or more objects. The graph is drawn using vertices and edges; graph theory is widely implemented in the area of traffic signals. Traffic management plays an important role to avoid accident; consequently we have to regulate a traffic flow. In this paper we express traffic flow using Pythagorean fuzzy graph. In fig.1, the traffic flow is defined by vertices and edges; the arrow represents the direction of vehicles. The number of vehicles passing in all paths is not similar. Since every left turn does not interfere with the traffic flow (i.e.) the vehicles can pass without any disturbance. Remaining direction in figure 1is marked from A to H indicate the traffic flow that met with an accident. The traffic flows are represented using Pythagorean fuzzy graph. In which the membership is denoted by the number of vehicles moving in the path. When number of vehicles passing in any path is high then membership value is consider to be a higher, when number of vehicles passing in any path is less then membership value is consider to be a lower, and non-membership is denoted by the waiting time of vehicles in the path.

Accident region depends upon the number of vehicles passing in each lane. So we have to consider a traffic pattern in which vehicles can pass through intersecting line without obstructing the other vehicles. In this problem, we denote each traffic flow using vertices of Pythagorean fuzzy graph. The traffic signal plays an important role to avoid vehicle collision. In given problem vehicle collision depends on vertices that are adjacent. For example, let us consider the direction of traffic flow from fig.1; in which A and H are coincide, so there is a chance of collision between vehicles. The adjacent vertices for A are C, F, G and H. Similarly the adjacent vertices for H are A, B, C, E and F It gives an idea that possibility of accident depend upon the adjacent vertices and its membership value. The number of vehicle is high there is a risk of traffic jam and accident. The vehicle passing is between the ranges 5,000 to 10,000 per hour; then its membership value is represented as higher, when vehicle passing is between the ranges 0 to 5,000 per hour. If the speed of vehicles is high there is a chance for accident. So speed limit for vehicles must be controlled in all paths in order to avoid collision of vehicles. The non-membership are represented using the waiting time of a vehicle in path, if waiting time in any path is high there will be risk for high traffic jam.

Traffic Signal Control
Consider the above road map with intersecting road; where signals are essential to take diversion in road to avoid vehicle collision. In fig.3, A to H represents the road. There is a free left in which vehicles can pass smoothly, thus every left turn does not require a signal i.e. $H \rightarrow F, E \rightarrow C, D \rightarrow B, A \rightarrow G$ where $\rightarrow$ represent the direction of vehicles in path. The table below indicate how signals are used in order to avoid traffic jam and accident. Thus signals play a great role in area of traffic control.

Numerical example
Consider a Pythagorean fuzzy graph Fig.1 with vertex set $\alpha = $ {A,B,C,D,E,F,G, H} and edge set $\beta = $ {BC,BD,BE,BH,CA,CH,CE,DG,DF,DE,EG,EH,FA,FG,FH,GA,HA}

Let a family of spherical fuzzy set be $\Gamma^* = \left\{c_1, c_2, c_3, c_4\right\}$ defined on X as:

$$c_1(v_i) = \begin{cases} (0.2, 0.5) & if\ i = A \\ (0.6, 0.8) & if\ i = E \\ (0,1) & otherwise \end{cases}$$

$$c_2(v_i) = \begin{cases} (0.3, 0.7) & if\ i = B \\ (0.7, 0.3) & if\ i = F \\ (0,1) & otherwise \end{cases}$$

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$$c_3(v_i) = \begin{cases} (0.1, 0.4) & if\ i = C \\ (0.6, 0.2) & if\ i = G \\ (0,1) & otherwise \end{cases}$$

$$c_4(v_i) = \begin{cases} (0.5, 0.3) & if\ i = D \\ (0.3, 0.4) & if\ i = H \\ (0,1) & otherwise \end{cases}$$

Hence the family $\Gamma^* = \{c_1, c_2, c_3, c_4\}$ satisfies the condition of Pythagorean fuzzy vertex coloring of the graph G. Therefore, the chromatic number is $\chi(G) = 4$. Thus, minimal number of way of coloring the graph is four. Thus graph G is called properly colored graph.

CONCLUSION

A Pythagorean fuzzy graph helps to find out the vagueness of problem in an accurate way. In this article, the traffic flows are represented by vertices and edges of Pythagorean fuzzy graph. Membership value represents the accidental zone in a city and non-membership value helps to analyse waiting time of the vehicles in all path. This paper provides an attempt to explain traffic flow using Pythagorean fuzzy graph, efficient way for planning of traffic signal and also helps to analyse about the minimal ways required for coloring the graph with an illustrative example.

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Table 1: Signal Control for Road H

GREEN SIGNAL – 1	RED SIGNAL – 1
Road – H **Vehicles allowed** $H \to C$ and $H \to B$	Vehicles not allowed $E \to B, E \to G$ $D \to F, D \to G$ $A \to F, A \to C$

Table 2: Signal Control for Road E

GREEN SIGNAL – 2	RED SIGNAL – 2
Road – E **Vehicles allowed** $E \to B$ and $E \to G$	Vehicles not allowed $D \to F, D \to G$ $A \to F, A \to C$ $H \to C, H \to F$

Table 3: Signal Control for Road D

GREEN SIGNAL – 3	RED SIGNAL – 3
Road – D **Vehicles allowed** $D \to G$ and $D \to E$	Vehicles not allowed $A \to F, A \to C$ $E \to B, E \to G$ $H \to C, H \to B$

Table 4: Signal Control for Road A

GREEN SIGNAL – 4	RED SIGNAL – 4
Road – A **Vehicles allowed** $A \to F$ and $A \to C$	Vehicles not allowed $D \to F, D \to G$ $E \to A, E \to G$ $H \to C, H \to B$

Table 5: Value of Vertex in Pythagorean Fuzzy Graph

| Vertex	A	B	C	D	E	F	G	H
Value	(0.2,0.5)	(0.3,0.7)	(0.1,0.4)	(0.5,0.3)	(0.6,0.8)	(0.7,0.3)	(0.6,0.2)	(0.3,0.4)

Table 6: Value of Edge in Pythagorean Fuzzy Graph

Edge	BC	BD	BE	BH	CA
Value	(0.1,0.7)	(0.3,0.7)	(0.3,0.8)	(0.3,0.7)	(0.1,0.5)
CH	**CE**	**DG**	**DF**	**DE**	**EG**
(0.3,0.4)	(0.1,0.8)	(0.5,0.3)	(0.5,0.3)	(0.5,0.8)	(0.6,0.8)
EH	**FA**	**FG**	**FH**	**GA**	**HA**
(0.6,0.8)	(0.2,0.5)	(0.6,0.3)	(0.3,0.4)	(0.2,0.5)	(0.2,0.5)

Table 7: Adjacent Vertices of Pythagorean Fuzzy Graph

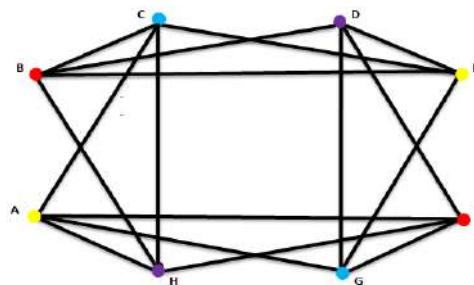| Vertex	Adjacent vertices
A	C,F,G,H
B	C,D,E,H
C	A,B,E,H
D	B,E,F,G
E	B,C,D,G,H
F	A,D,G,H
G	A,D,E,F
H	A,B,C,E,F
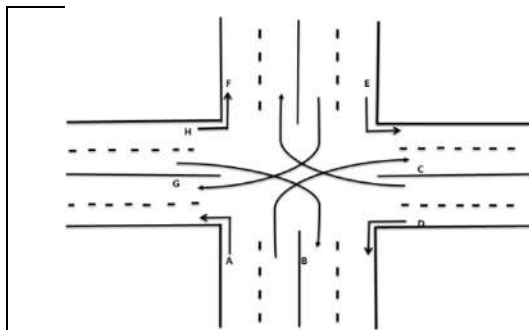
Fig 1. Road Map
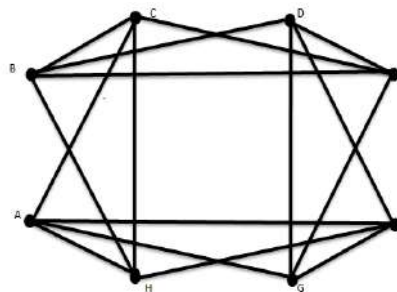
Fig 2. Pythagorean Fuzzy Graph
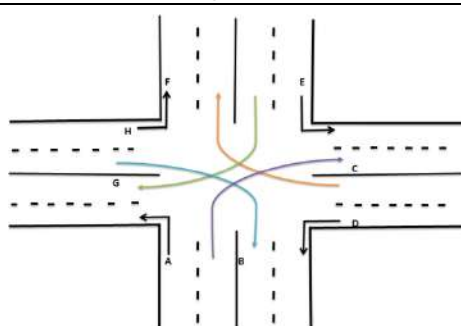
Fig 3. Road Map for Signal Control

Fig 4. Vertex Coloring of Pythagorean Fuzzy Graph

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REVIEW ARTICLE

An Overview on Advanced *In-vivo* and *In-vitro* Models for Investigation of Nephroprotective Potential of Drugs

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ABSTRACT

The kidney is an important major target organfor evaluation of drug-induced toxicity. Use of many therapeutic drugs is a common source of acute kidney injury. The use of such drugs may produce nephrotoxicity is a major problem of kidney dysfunction which may affects potential kidney functions and can affect the investigation of new herbal drugs and pharmaceutical products. To control drug induced toxicity to the kidney the new methods are needed for evaluation of herbal crude drugs &chemically synthesized drug. In this review article we have described overview of methods which are being used to induce nephrotoxi city (in-vivo as well as in-vitro methods)for evaluation of curative or protective effect of drugs. The objective of this review paper is to provide the collective list of most relevant models for in-vivo & in-vitro neuroprotective evaluation. Here we provided collective information of different *in-vivo* and *in-vitro* models which are being utilized over the last decades to evaluate drug induced renal injury which will serve as a useful resource to understand the mechanism of in vivo & in vitro models for nephrotoxicity. The brief information of all these methods we have explained which would be helpful for more sophisticated, precise, effective response of drug to minimize the use of animals and could be more relevant model for researcher for nephroprotective evaluation of drugs.

Key words: Drug induced toxicity, nephroprotective, *in-vivo* models, *in vitro* models

INTRODUCTION

The kidney is an essential and prime organ in human body, which receives an abundant blood flow of 20-25% of cardiac output and removes xenobiotic and metabolic products from the blood into the urine (Sales & Foresto, 2020). There are threeprocesses are involved during the formation of urine in kidneys, including glomerular filtration, tubular reabsorption and tubular secretion (*Pharmacology behind Common Drug Nephrotoxicities*, n.d.). The glomerular filtration is essential process for the kidneys to rapidly remove waste products and toxins from the blood. The renal tubules are responsible for reabsorption and secretion of substances (H. Wu & Huang, 2018). The different compartment of kidneys are normally exposed to high concentrations of metabolites as well as drugs, therefore, causes susceptibility to drug toxicity (H. Wu & Huang, 2018). Drug-induced nephrotoxicity is one of the major pathogenic factors of acute kidney injury (AKI), chronic kidney disease (CKD), acute renal failure (ARF) and end-stage renal disease (ESRD).(Coca et al., 2012)It was predicated that drug-induced renal failure is accounted for 25% of all cases of acute renal failure (Lopez-Novoa et al., 2011), the incidence in older patients is even as high as 60%.(H. Wu & Huang, 2018).

The diagnosis of drug-induced nephrotoxicity at an early stage is difficult to determine, because slight renal damage and mild urinary sediment abnormalities that associated with some prescribed medications are frequently unrecognized. In consequence, detection of nephrotoxicity is often delayed until an overt change in renal function with an increasing level in serum creatinine and an early therapeutic opportunity is generally lost. Drug-induced renal injury may be resulted from cumulative dose-dependent toxicity or idiosyncratic dose-independent toxicity at any time during therapy. (Basile et al., 2012)In the past decade, extensive efforts have been made to identify serum and urine biomarkers for diagnosing renal cellular injury, such potential biomarkers including kidney injury molecule-1 (KIM-1), neutrophil gelatinase-associated lipocalin (NGAL), clusterin and urinary proteins with enzymatic activity like N-acetyl--glucosaminidase (NAG). Some cases of renal injury can be predicted and monitored. For example, cumulative dose-dependent renal toxicity can be anticipated and prevented, but idiosyncratic renal toxicity is difficult. (Bonventre, 2009), (Mishra et al., 2003)Therefore, understanding the mechanisms and biomarkers of drug-induced nephrotoxicity and exploring the strategies to prevent drug-induced nephrotoxicity could predict and detect kidney injury at an earlier stage, as well as decrease the risk of renal failure. In this review, simplified brief information on drug-induced kidney injury mechanisms with available and recently used *in-vivo* & *vitro*-models to predict effective relevant models for nephroprotective activity is discussed, with aspecial focus on novel developments in the field of investigation of nephroprotoxic drugs.

Drug-induced kidney injury:
Nephrotoxicity induced by drug is a type of injury which may cause directly or indirectly by the use of medications. The toxicity of kidney varies from an acute or chronic lowered glomerular filtration rate (GFR) to nephritic syndrome and renal tubular damage.(Sales & Foresto, 2020). Nephrotoxicity by toxins may produce acute or chronic kidney failure or physiological injurymainly due to of peculiar injury to tubular epithelial cells. The common pathways of cytotoxic injury are associated with final ischemic-reperfusion injury which lead to cellular adaptation, low toxic or toxic injury, and activation of regeneration and repair pathways. (Walker & Endre, 2013)

Mechanism of nephrotoxicity
A mechanism of nephrotoxicity drug-induced nephrotoxicity is classified as either dose dependent or dose independent. (Downes et al., 2020), (Downes et al., 2020) Prediction of dose-dependent toxicities and it may be correlated to the main pharmacological effect of the drug (type A reactions). Dose-dependent toxicities can be warding off by reducing dose, however sometimes itis necessary fortermination of therapy. Dose-independent toxicities, known as type B reactions, are genetically determined, may occur at any time during therapy and are highly varying from patient to patient.(Downes et al., 2020), (Fuchs & Hewitt, 2011)

Models for nephrotoxicity screening
Antimicrobial drug induced nephrotoxicity

Antimicrobial agents are one of the most commonly prescribed drug classes in children. In a clinical study was done in 2012, and suggested that the use of antimicrobial agents can develop nephrotoxicity in 37% of hospitalized children across in different hospitals were receiving antimicrobials including 61% of pediatric ICU, patients.(Versporten et al., 2016)Study has investigated and reported that lifesaving and often critical, many antibiotics are unfortunately produced nephrotoxic effects. In other study scientist observed and explained that, many class of antimicrobials have potential to cause nephrotoxicity, (Choudhury & Ahmed, 2006) and the frequency of toxicity may vary which depends on the properties of the individual drug, as well as the physiological status and underlying condition of the patient receiving the drug. (Hofmann et al., n.d.)There are variety of primary nephrotoxicity like tubuloepithelial injury acute tubular necrosis (ATN), also tubulointerstitial disease acute interstitial nephritis (AIN) and obstructive crystal nephropathy was developed by antimicrobial drugs. (Downes et al., 2020)

Antibacterial Models
Aminoglycosides model

It has been reported in the research study that, one the most useful class of antimicrobial agent i.e. aminoglycosides are nephrotoxic may develop a dose-dependent decrease inof kidney function of patients.18(Moore et al., n.d.)Mostly common cause of aminoglycoside-induced renal toxicity is due to toxicity to proximal convoluted tubule. Then aminoglycoside entersthe cytosol and accumulates in organelles such as themitochondria, Golgi complex, and the nucleus.(Nagai & Takano, 2004)In few cases of distal tubule damagewith hypomagnesaemia and decreased concentrating abilityhas been reported.(Patei & Savage, 1979)In this aminoglycoside category, gentamicin is antimicrobial agent is the most nephrotoxic drug which commonly used, followed by tobramycin and amikacin. Among the aminoglycosides neomycin is high nephrotoxic, neurotoxic and ototoxic potentialas it has highest affinity to proximal convoluted tubule(PCT) and therefore is rarely used clinically.

In-vivo Model and Experimental Protocols
Gentamicin model

The nephrotoxicity can be induced using 80 mg/kg volume (5 mL/kg) gentamicin administered in animals by intraperitoneal injection and toxicity developed in rats by administration of drug once a daily.(Jado et al., 2020) Mechanism behind the gentamicin-induced toxicity to kidney in rats may be associated as hyper cellularity occurs in mesangial cells and association of parietal and visceral layers of Bowman's capsule (proliferative glomerulonephritis; arrow), with degeneration of renal tubular epithelium. (Abdelrahman, 2018). The use of aminoglycosides antimicrobials models for nephroprotective activity is most popular method the gentamicin-induced of herbal drugs; however newer method could be adopted for induction of nephrotoxicity. The nephrotoxicity induced by gentamicin in rats had been reported that, oxidative damage with significantly decreasing creatinine, SOD, LDH, BUN, protein in urine & urine volume. (Patei & Savage, 1979)In addition to this researcher investigated and observed that, gentamicin induced nephrotoxicity may alter histopathological parameters with elevation of in the biomarkers like serum level of Creatinine, Blood Urea Nitrogen (BUN), renal neutrophil gelatinase-associated lipocalin (NGAL), and Kidney injury molecule-1(KIM-1). In other study biomarkers of nephrotoxicity which increases due to gentamicin induced nephrotoxicity like malondialdehyde (MDA), protein carbonyl (PC), and nitric oxide (NO)• level; as well as decreased cellular antioxidant activity of glutathione (GSH) content and superoxide dismutase (SOD) and catalase (CAT) activity in the kidney tissues. (Amin, 2021)

Amikacin model

The other antimicrobials like amikacin can be used as an animalmodel in animals by inducing nephrotoxicity with that of renal toxicity in humans. The researcher investigated by subcutaneous administration of amikacin with single-dose of 100 or 500 mg/kg of body weight in Sprague-Dawley rats.(Chan et al., 2020)The satisfactory results by amikacin induced toxicity in nephrons may be useful for induction of nephrotoxicity in animals it may be associated with a higher amikacin concentration in renal tissues which can be correlated with the study protocol.(Chan et al.,

2020), (Kaynar et al., 2007), (Kara et al., 2016) The studies has been done on the impact of amikacin induced nephrotoxicity rats and have reported that, increase in reactive oxygen radicals species (ROS) responsible for lipid peroxidation which develops malondialdehyde (MDA)which may develop nephrotoxicity. In addition to this more lesions with glomerular congestion, mononuclear cell infiltration, tubular dilatation, cortical and medullary hemorrhage, proximal and distal tubular degeneration, which indicate renal tissue damage.(Asci et al., 2015)

Adriamycin model

In previous research study it was reported that anephritic syndrome can be developed by use of Adriamycin (10 mg/kg)injection by tail-vein and evaluation were done in that the structure and ultrastructure alteration, podocytes proteins expression, was investigated for heparinase activity.(Assady et al., 2015)The mechanism of development of nephrotoxicity by Adriamycin is due to direct toxicity and damaging to the glomeruli along with injury to tubular interstitial tissue. Adriamycin causes changes to the barrier of glomerular filtration, including endothelial cells of glomeruli, basement membrane of glomeruli and podocytes. (V. W. Lee & Harris, 2011). This model can be used to induce nephrotoxicity by Adriamycin in the nephroprotective investigation of drugs.

Rifampicin model

The use of rifampicin most effective drug treatment in TB from aminoglycosides category have reported for nephrotoxicity and have renal toxicity by 0.8 ml/Kg injection intraperitonealto induce nephrotoxicity. Investigators observed that the administration of Rifampin in animal with a dose of 50 mg/kg body weightgiven orally.(*Ruta Graveolens Protects Against Isoniazid/Rifampicin-Induced Nephrotoxicity through Modulation of Oxidative Stress and Inflammation*, n.d.)This dose causes renal injury and glomerular dysfunction with elevation of serum urea, creatinine, sodium and potassium levels act as nephrotoxic drug. (Ramadan et al., 2016). In addition to this cause lesions including atrophy of glomerular tuft, dysplastic renal tubules and inflammatory cells infiltration (*Ruta Graveolens Protects Against Isoniazid/Rifampicin-Induced Nephrotoxicity through Modulation of Oxidative Stress and Inflammation*, n.d.)The use of rifampicin for induction of nephrotoxicity would be alternative model in assessment nephroprotective effect of investigating drugs.

Vancomycin model

The current use of antimicrobial agent vancomycin may be effective model for development of nephrotoxicity in animals with creating variety of risk factors for Acute Kidney Injury (AKI). The much more commonly Vancomycin Associated Nephrotoxicity (VANT) occurs, thebased on some experimental studies it was reported that, the toxicity of vancomycin to kidney is due to the development of tubular ischemia by oxidative stress. In recent studies,demonstrated that, the vancomycin nephrotoxicity due to formation of vancomycin cylinders with uromodulin, with greater attention in plasma values, and early increased creatinine level compared with other nephrotoxic drugs. (Zrouri et al., 2021)The use of two combined combined use of piperacillin-tazobactam (PTZ) with vancomycin was reported as enhanced potential nephrotoxicity compared to vancomycin without this combination.(Filipponeet al., 2017).

Antiviral nephrotoxicity
Oral acyclovir model

In the clinical study have observed that, the use acyclovir can cause changes of retinol-binding protein and β - microglobulin, indicating injury to the proximal renal tubule and acute renal failure (ARF) in severe acute pancreatitis (SAP). The ARF was due to SAP and have observed and mentioned that clinical manifestations including increased levels of blood, urine amylase, serum creatinine, blood urea nitrogen abdominal pain, with slight swelling of the pancreas. (Meng et al., 2011)The use of acyclovir antiviral drug could be considered as good model for nephrotoxicity in rats as an alternative method in development of kidney injury.

Tenofovir model

It has been investigated & reported that, the use of antiviral drug tenofoviras active metabolite which may be responsible to produce the by altering DNA expression of endothelial nitric oxide synthase due to accumulation of

tenofovir in renal proximal tubular cell, which may cause dysfunction of sodium-phosphorus cotransporter, sodium/ hydrogen exchanger 3, and aquaporin 2. The formation of disoproxil fumarate (TDF) after administration of tenofovir and exposed to renal tissues which may increase risk of proteinuria, increased risk of rapid decline, and increased risk of chronic kidney disease. (Novick et al., 2017)

Antifungals model
Nephrotoxicity can be induced by the use of antifungal agents which is having major treatment complication associated with the treatment of fungal infection.(Udawatte et al., 2020) Amphotericin is most preferably used antifungal agent which has dose-dependent mechanism-based potential for glomerular and tubular toxicity and which can induce nephrotoxicity insexisting renal damage, immunocompromisedor severely ill patients with collateral therapies with other potentially nephrotoxic drugs.(Tragiannidis et al., 2021). The Mechanism behind the development of nephrotoxicity by amphotericin is due to direct action on the renal tubules as well as of drug-induced renal vasoconstriction with a subsequent reduction in renal blood flow and glomerular filtration rate. (Fanos & Cataldi, 2000)

Antineoplastic drugs
Cyclophosphamide model
The commonly used alkylating antineoplastic drug which may develop nephrotoxicity can be an effective model for nephroprotective potential effect of drugs. The commonly use antineoplastic drug like cyclophosphamide renal toxicity can be developed within 7 days at dose of 150 mg/kg body weight using intraperitoneal route in rat model. (Mahipal & Pawar, 2017)The mechanism behind the toxicity in nephrons is due to increased production of both reactive oxygen and nitrogen species in kidney tissues, along with the lipid peroxidation and inhibition of superoxide dismutase activity.(Ayza et al., 2020)(Ayza et al., 2020)

Cisplatin model
The alkylating antineoplastic agent cisplatin induced soxidative stress and inflammation in animals may be the mechanisms of cisplatin-induced nephrotoxicity and suppressed the endogenous antioxidant machinery in renal system. (Ibrahim et al., 2018)Cisplatin induced nephrotoxicity is indicated with elevated levels of renal biomarkers like serum transaminases, oxidative stress markers andurine kidney function markers, and histopathological changes in kidney.(Kpemissi et al., 2019)The based on reported study use of high dose & short duration of cisplatin can be used for induction of nephrotoxicity inanimal models for most common and severe side effects i.e. acute kidney injury (AKI) by cisplatin in can patient.(J. Lee et al., 2020) The cancerous model with the evaluation of nephroprotective action can be investigated together by development of such model at laboratory level.

Methotrexate (MTX) model
The use of antimetabolite and antifolate antineoplastic drug methotrexate which can cause renal injury induced by use of 20mg/kg i.p. or 60mg/kg/orally is mainly associated with its accumulationof drug and along with its metabolite in the kidney tubules. A significant rise in serum creatinine and blood urea nitrogen (BUN) was reported in the study. The use of methotrexate indicated that animpairment of renal function may be responsible for toxicity in renal tissues.(Sherif et al., 2019)It is mentioned and suggested that methotrexate produces more than twice higher levels of serum creatinine, cystatin C, and BUN, along with more than ten times higher level of microalbuminuria, which is responsible for development of a significant renal impairment in the animal model.(Abouelela et al., 2020)

Ifosfamide Model
The use of another an alternative cytotoxic a prodrug Ifosfamide is nephrotoxic compound i.e. chloroacetaldehyde (CAA) which can be effective method for induction of nephrotoxicity in rats.(Choucha-Snouber et al., 2013). Based on the pharmacokinetic study, it is reported that the toxicity by ifosamide is due to development of metabolizing agent chloroacetaldehyde (CAA) on renal cells via its (IFO) by liver cells. This model described the production of CAA by the liver cells and its transport to the renal cells can be considered for an effective alternative model.(Leclerc et al., 2016)

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Carfilzomib (CFZ) model

The use of an effective non-reversible selective proteasome inhibitor anticancer drug carfilzomib (CFZ) is usedin myeloma. (Fotiou et al., 2020)The mechanisms of carfilzomib caused renal injury by may be due to prerenal damage, with prerenal or vascular short-term rise in creatinine, observed during the therapy of carfilzomib. So this antineoplastic agent induced nephrotoxicity can be an effective model in pathophysiology of cancer for testing of nephroprotective activity of drugs.(Jhaveri et al., 2013)

Immunosuppressive model
Cyclosporine A (CsA) model

Cyclosporine is used to treat psoriasis and immunosuppressive agent which can elicitan effective nephrotoxicity, with increased serum levels of creatinine and BUN classical nephrotoxicity markers, alongside an increased renal NGAL protein expression. (Arab et al., 2021)In the previous study suggested that, cyclosporine can generate nephrotoxicity by its direct effects on renal tubular epithelial cells which include the induction of epithelial-mesenchymal transition (EMT). (Q. Wu et al., 2018)The cyclosporine can be administered in a daily 15 mg/kg/day dose by subcutaneous route for induction of nephrotoxicity(Carlos et al., 2014). The use of cyclosporine treatment of 75 mg/kg daily for 4 weeks in mice can develop severe nephrotoxicity with histopathological changes like tubular vacuolization, casts, and cysts in renal tissues.(Ramírez-Bajo et al., 2020)

Tacrolimus model

The most used and effective but limited use immunosuppressive agents tacrolimus produces secondary effect i.e., nephrotoxicity in the treatment of transplantation. The mechanisms of nephrotoxicity remain unknown. but may be related to elevation of the chemokine receptor which enhances the renal fibrosis progression which develop toxicity in renal tissues. (Wang et al., 2019)The intraperitoneal injection of tacrolimus at 2 mg/kg per day can develop toxicitywith various side effects, and most common is nephrotoxicity can developed in animal group. (Wang et al., 2019).Researcher suggested that, the drug-induced nephrotoxicity is the major dose-limiting side effect of tacrolimus which limit its use. (Alshamsan et al., 2020)

NSAIDs Induced Nephrotoxicity model
Acetaminophen (AAP) model

Acetaminophen is most preferably used and easily available OTC analgesics& antipyretic drug. Use of acetaminophen causes nephrotoxicity by the toxic effect of N-acetyl-p-benzoquinone imine (NAPQI) by oxidation in liver. The development of acetaminophen induced toxicity by formation of reactive oxygen species (ROS) causes nephrotoxicity in renal tissues.(Chinnappan et al., 2019)The acetaminophen induced nephrotoxicity model can be developed by intraperitoneal administration of 200 mg/kg dose per day for 14 days. The mechanism of nephrotoxicity by acetaminophen may be due to tubular injury in the kidneys and with appearance of markers in urine like phosphaturia, low molecular weight proteinuria which may turn into severe renal failure.(Neelima et al., 2020). In addition to this, an elevation of urea and creatinine in animal group provided significant results for kidney injury. (Chinnappan et al., 2019)

Diclofenac model

Diclofenac is most prescribed and recommended drug for pain and inflammation which may develop the toxicity in renal system of rats with significant elevation of serum creatinine and urea is the mechanism for diclofenac induced development of nephritis& nephrotoxicity. In addition to that, reduction of prostaglandin E-2 (PGE-2) production due to COX-2 inhibition could be the result for renal damage.(Mostafa et al., 2020)The necrotic changes indicated at a 50 mg/kg dose rate, which develop oxidative stress in the kidneys. In addition to this pyknotic nucleus develops in the tubular epithelial cells in the renal parenchyma of the animals (rabbit).(Iftikhar et al., 2015), In other studies it has been reported that, for the development of nephrotoxicity diclofenac sodium was administered with the dose of 100 mg/kg body weight/day, i.p. for evaluation of nephroprotective effect of drug.(Famurewa et al., 2020). The treatment of diclofenac in mouse causes acute tubular necrosis in the kidney, including development of vacuole in the tubular

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epithelium, sloughing of tubular cells into the lumen, tubular dilation and loss of brush border with depletion of the antioxidant defense molecules glutathione (GSH) and superoxide dismutase (SOD).(Huo et al., 2020).

Radiographic contrast media (RCM) induced nephrotoxicity

The third leading cause of hospital-acquired acute renal failure most commonly due to use of radiographic contrast media (RCM) in diagnosis and interventional procedures. (Karaman et al., 2016)Contrast media-induced (CIN) nephropathy (CIN) is caused by the iodinated contrast media acute kidney injury (AKI) within 48 hours to 72 hours after CM administration. The primary mechanisms by which CM causes AKI are by causing renal ischemia and by direct tubular epithelial cell toxicity. (Brezis, 2005), (Lohani & Rudnick, 2020)It has been reported that, the nephrotoxicity induced by single dose of 10 ml/kg body weight by intravenous administration via tail vein of iohexol (contrast media, CM) over a period of2 min to the animals.

Proton pump inhibitor induced model
Omeprazole model

The proton pump inhibitors (PPI) are most commonly prescribed inpeptic ulcer and gastroesophageal a reflux disease. One of them is omeprazole is self-medicatingin significant percentage PPIs are among the most commonly prescribed drugs.(De Milito et al., 2007)Based on previous study, the use of omeprazole in mice at dose of 40 mg/kg/day administered intraperitoneal route for 10 or 28 days of study.(De Milito et al., 2007)The mechanism of nephrotoxicity by PPI is either due to interference in lysosomal acidification, proteostasis or may be due to hypomagnesemia increased oxidative stress, with acceleration of wear and tear damage in human renal endothelial cells.(Arif, 2017)With structural damage and loss of function of renal system and significant appearance of renal biomarkers for drug-induced kidney injury in conventional in vivo model which may be the good models for testing of nephroprotective action of drug. (Cunningham et al., 2010). The in-vitro methods are poorly predictive for nephrotoxicity in animal models or humans.(Magee et al., 2013). However, the use of such in-vitro studies could be effective in minimization of methodological error, minimum usage of animal with more productive results. (Huang et al., 2015)Some of the advanced methods of in-vitro methods can be beneficial for more economical, accurate prediction and minimization of use of animal for nephroprotective screening of new chemicals.

Advanced *in vitro* models

In vitro methods offer rapid and cost effective screening methods in specific types of cell for specific effects. (Choudhury & Ahmed, 2006), Hofmann et al., n.d.) In vitro screening method provides platform for evaluation of chemical to differentiate direct and indirect effects at a cellular and subcellular level. In vitro methods have been played crucial role in helping to understand the mechanisms of well-developed nephrotoxins.. (*In Vitro* Methods in Pharmaceutical Research by Jose V. Castell, Maria Jose Gmez-Lechn (z-Lib.Org), n.d.) Recently, to minimize the mortality and use of laboratory animals in research study, the use vitro modelsas primary cells are with similar the physiological action of cells likes to that in vivo. Despite of low growing capacity, to study basic renal cellular functions and the effects of nephrotoxic drugs primary renal cells are still remaining a most effective option. (Faria et al., 2019)

Common sources of kidney cells

Primary renal cell cultures provide a more realistic model with abundance of renal transporters, with the exceptions of BCRP and MATE-2K, within the human kidney cortex has been quantified (Prasad et al., 2016).The use of kidney cell lines to evaluate transporter interactions in the context of nephrotoxicity have been previously reviewed (Fisel et al., 2014; George et al., 2017).The kidney proximal tubule epithelial cell (PTEC) is the most common cell type used for in vitro evaluation of kidney toxicity. Single-layer two-dimensional (2D) cultures of kidney PTECs from various origins have been used to investigate the toxic effect of drugs on the kidney.(Nieskens & Sjögren, 2019) Freshly isolated primary human proximal tubule epithelial cells (HPTECs) most reliable in vivo cells in terms of morphology, polarization, drug transporter activity, and biomarker expression. (Nieskens & Sjögren, 2019)

Novel *in-vitro* models for nephrotoxicity:
Kidney on chip
Rodents are used continuously in preclinical studies but has some drawbacks of at the time of evaluation of nephroprotective efficacy of drugs which are the nephrotoxic drugs have severe toxicity to kidney. (Barnett & Cummings, 2018). It was suggested the use of kidney-on-a-chip could be the effective method for prediction of assessment of nephrotoxic and nephroprotective drugs which will be helpful minimize the gap between traditional *in-vivo* method could be helpful for prediction of mechanism of action.(Wilmer et al., 2016)

Pluripotent stem cells
Use of stem cell–derived in vitro models can be adopted that use of 3D multicellular kidney models which derived from hPSC (human pluripotent stem cell), can minimize the severity of nephrotoxins from benign compounds.(Bajaj et al., 2018)

Organoid
In the future use of use of organoid could be more effective and predictive vitro model, in which the very complex structures of kidneys can be replaced which can produce with same efficacy of kidney like blood filtration and urine re-absorption units could be developed for their function and homeostasis.(Soares et al., 2020)

Bio functionalized hollow fibers
In the research study investigated that immortalized proximal tubule epithelial cells can be utilized on the surface of hollow fibers of exterior part which can be coated with an extracellular matrix, making such a monolayer of cells biofunctionalized hollow fibers could be useful as in vitro model. This could be emerging method of in vitro model can be useful in future research with minimization of toxicity to animal and to obtain more predictive and with time limits method. (Soo et al., 2018)

CONCLUSION

In kidney nephrotoxicity may be associated with notable death and development of disease conditions. Currently the drug-induced nephrotoxicity is a problem with impairments of renal system there is a need to develop more effective kidney-based in-vivo and in-vitro model for the study of nephrotoxicity. Nowadays, vast majority of drugs, chemicals, herbal plants are being used globally used to investigate the potential of test compound against nephrotoxicity. The knowledge of the mechanisms of kidney injury, the drugs that originate it, the risk factors for its development, and the methods for its prevention and/or treatment, is required. In this review we focused on useful information of drugs used clinically and have good potentials for development of nephrotoxicity. This study provides some evidence against development of nephrotoxicity in both in vitro as well as in vivo evaluation of drugs. The information obtained from these in-vivo & vitro models which will prevents the unnecessary wastage of drugs from the development pipeline but also ease the drug development of safe drugs and nephrotoxic adverse effects can be managed more effectively in clinical use. Hence, this review concluded that the many animal models can be developed and used for renoprotective potential of many herbals, synthetic drugs in future preclinical studies.

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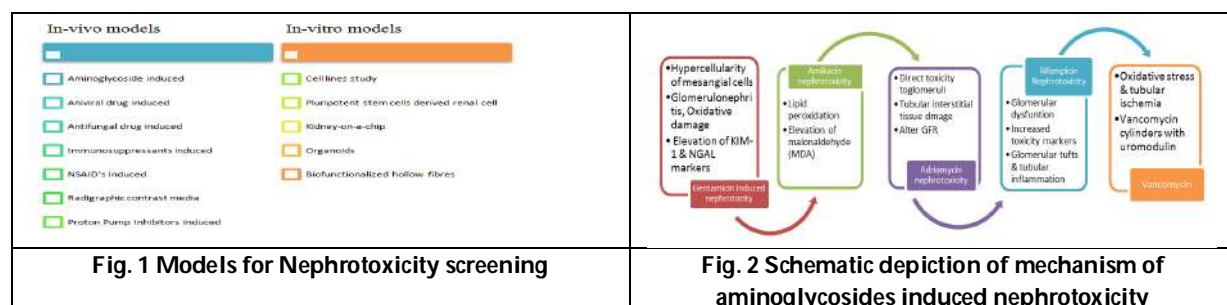
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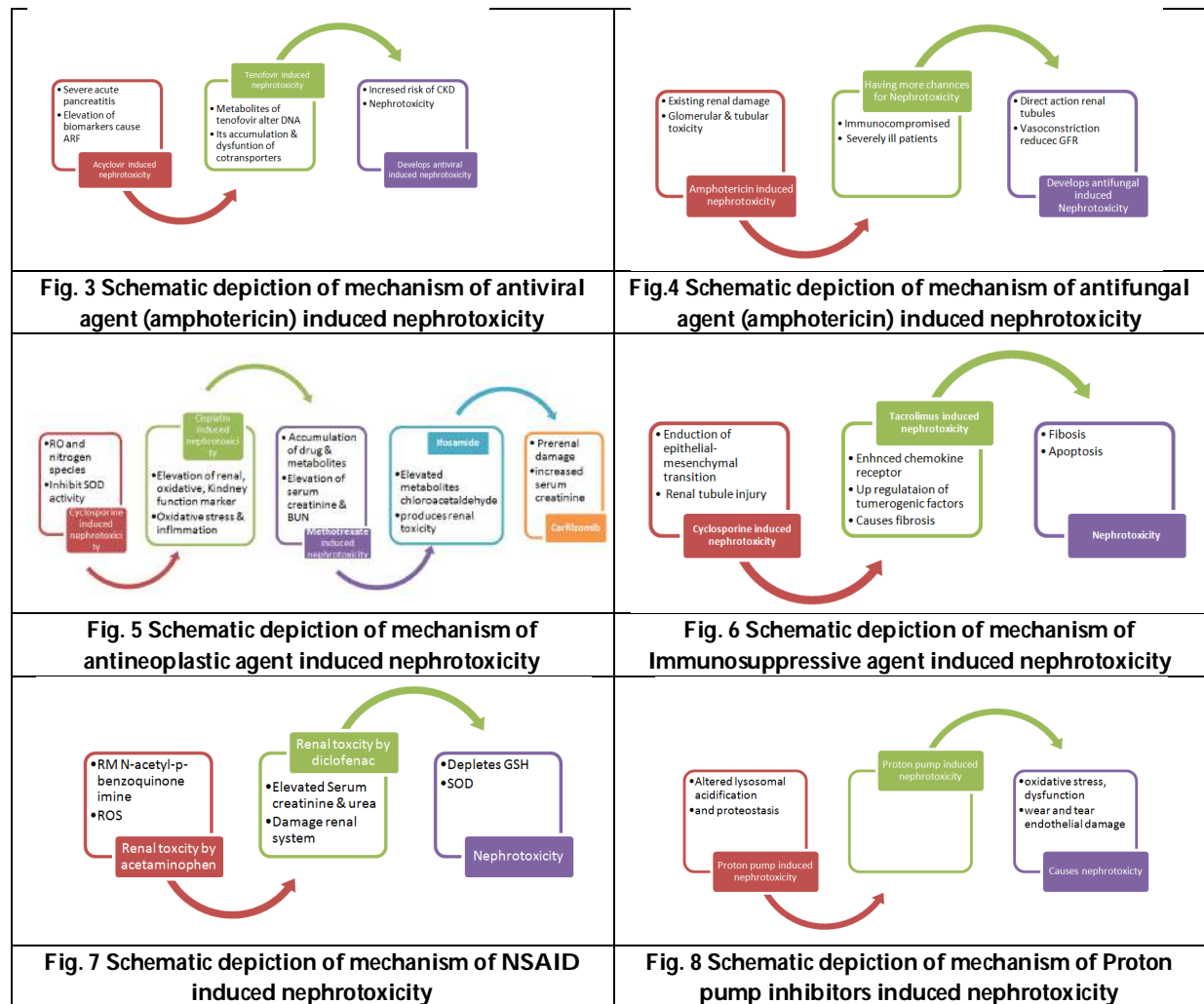
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| Fig. 1 Models for Nephrotoxicity screening | Fig. 2 Schematic depiction of mechanism of aminoglycosides induced nephrotoxicity |

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Fig. 3 Schematic depiction of mechanism of antiviral agent (amphotericin) induced nephrotoxicity

Fig.4 Schematic depiction of mechanism of antifungal agent (amphotericin) induced nephrotoxicity

Fig. 5 Schematic depiction of mechanism of antineoplastic agent induced nephrotoxicity

Fig. 6 Schematic depiction of mechanism of Immunosuppressive agent induced nephrotoxicity

Fig. 7 Schematic depiction of mechanism of NSAID induced nephrotoxicity

Fig. 8 Schematic depiction of mechanism of Proton pump inhibitors induced nephrotoxicity

RESEARCH ARTICLE

In Silico Design and Molecular Docking Studies of Chalcone Derivatives As Topoisomerase II Inhibitors

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ABSTRACT

Cancer, is one of the leading cause of death world wide. Topoisomerase II is used as molecular target for developing novel inhibitors for cancer treatment. This study aims to identify Chalcone derivatives as Topoisomerase II inhibitors using in silico design and molecular docking studies. In silico design of proposed derivatives were conducted and derivatives obeying Lipinski's rule of five were selected for molecular docking studies by Auto Dock Vina using the protein ID5gwk from Protein data bank. Visualization and analysis were conducted by Py MOL. Molinspiration studies revealed that, the designed derivatives had physical and chemical properties meant for an orally bio available drug. Based on the docking results, derivatives PTC-3 and PTC-5 showed high affinity and high polar interaction towards active site of protein 5gwkwq. The designed Chalcone derivatives were found to possess good binding affinity and good interaction in the binding pocket of target 5gwk, so these derivatives are expected to exhibit good anticancer property in*in-vitro* and *in-vivo* studies.

Keywords: Cancer, Topoisomerase II, Docking, Auto Dock Vina, Etoposide.

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INTRODUCTION

Cancer, a generic term, which is used to refer a group of diseases that can affect any part of the body. It is also known by the name malignant tumor and neoplasm. It involves abnormal cell growth with the potential of spreading to other parts of body known as metastasis, which is the primary cause of death from cancer. It occurs as a result of the interaction between a person's genetic factors and external agents called as carcinogens. They are categorized into three main classes, physical, chemical and biological carcinogen. Physical carcinogen includes ultraviolet and ionizing radiations. Chemical carcinogen includes asbestos, arsenic, aflatoxin, components of tobacco smoke etc. Biological carcinogen include infections from certain viruses, parasites or bacteria's. Worldwide it is one of the leading causes of death. About 10 million people died due to cancer in 2020[1]. The most common cause of cancer deaths in 2020 were lung cancer (1.80 million death) followed by breast (6,85,000 deaths), stomach (7,69,000 deaths), colon and rectal (9,35,000 deaths) and liver cancer (8,30,000 deaths)[2]. A correct diagnosis is an essential factor for effective treatment because every cancer type requires a specific treatment regimen. Radiotherapy, chemotherapy and/ surgery are the common treatment methods. There are several drugs available for cancer treatment. Target therapy is one of such treatment methods which interfere with specific proteins that help tumor growth and spread. Both Small molecule drugs and monoclonal antibodies are used in targeted therapy[3].

Topoisomerase enzyme is one of the effective targets for cancer therapy and development of novel anticancer drug. It plays an important role in DNA replication especially in the over winding and under winding of DNA. This problem arises during replication and transcription when DNA get over wound ahead on replication fork. The torsion results in the arrest of DNA or RNA polymerases to continue down the strand. Such topological problems of DNA are corrected by DNA topoisomerases. Topoisomerases can be further classified into two families; type 1 and type 2. Type I family is again classified into two subfamilies type I a and type I B. [4] Topoisomerase targeting drugs mainly act through topoisomerase poisoning resulting in replication fork arrest and double strand breaking. Recent studies suggested that isoform specific human topoisomerase II inhibitors may be developed as safer anticancer agents. [5] Some of the most powerful anticancer drugs used clinically as topoisomerase inhibitors are Etoposide, Tenoposide, Daunorubicin, Mitoxanrone, Amiscarine etc. Natural compounds such as alkaloids, polyphenols, terpenoids and flavonoids were reported to exert anticancer activity by targeting various metabolic pathway [6]. Chalcone is an aromatic ketone and an enone that forms a central nucleus for a variety of biologically active compounds. They are generally known as chalcanoids., that comes under flavonoid family [7] It is a privileged scaffold in medicinal chemistry. Various synthetic compounds derived from chalcones such as chalcones attached to heterocyclic rings like pyrazole, indole etc are proved to be effective anticancer agents [8,9] They also demonstrate anti-inflammatory action [10].

In recent years *In silico* drug designing methods have been actively used to expand the knowledge on the structure and role of Topoisomerase II and development of novel inhibitiors [11]. In designing structure-based approaches such as docking and molecular dynamic simulations, ligand-based approaches such as QSAR were studied. Molecular docking is a kind of bioinformatics modelling which involves the interaction of two or more molecules to give a stable adduct [12]. The information obtained from docking can be used to suggest stability of drug-target complexes, binding energy as well as free energy[13] In the present study various chalcone derivatives designed and determined their interaction with topoisomerase II (PDB ID: 5gwk) by means of molecular docking studies.

MATERIALS AND METHODS

ACD Lab Chemsketch 12.00
Used for drawing chemical structures, 3D optimization and calculating various physicochemical properties of the proposed derivatives.

Molinspiration

It is used to calculate the molecular properties and bioactivity for prediction of Lipinski's rule of five. Lipinski's rule of five or rule of thumb helps to determine whether the compound is likely to have the physical and chemical properties to be orally bioavailable. 5 derivatives designed were analysed by molinspiration and those derivatives obeying Lipinski 'rule of 5 were selected for docking studies.

Protein Data Bank (PDB)

PDB is the only crystallographic database meant for obtaining the 3D structural data of large molecules such as proteins and nucleic acids. Figure 1: 3D structure of topoisomerase 2. The structure was generated after X- ray crystallography and NMR studies. In this study the protein selected is topoisomerase II (PDB code- 5gwk) [14].

Molecular Docking

Docking is the prediction of affinity and activity of derivatives to suitable protein targets. Auto Dock Vina, an open-source docking program was selected for docking studies. PyMOL was used for protein preparation and visualization. PyRx was used for docking analysis.

Protein Preparation

Structure taken from the PDB database could not fit as such for docking studies. Because it consists of water molecules (HOH), detergents (DSN), small molecules, cofactors, metal ions etc. Therefore, the PDB structure should be converted into suitable form for docking by addition of command "remove<>resn<> molecules" (HOH, DSN etc). Hydrogen atoms should be added to the protein structure.

Ligand Preparation

The structures of derivatives were drawn using ACD Lab Chemsketch 12.0 and converted into 3D PDB format using Corina online software.

Docking by AutoDock Vina

Docking was performed using PyRx by loading the protein and derivatives into the navigation pane. The protein was then converted into macromolecule and derivative was converted into ligand molecule. After preparation of protein and ligand, click the AutoDock Vina Wizard button and adjust the grid size. The accuracy of the result depends on the number of exhaustiveness. Exhaustiveness is the number of times the ligand must be docked against the protein in different positions. After the completion of process, the results are displayed in the table. The binding affinity of the protein is indicated in Kcal/mol [15].

Visualization and Analysis

The PyMOL molecular graphics system was used to analyse the hydrogen bond, hydrophobic and pi-pi interactions. PyMOL can produce high quality 3D images of small molecules and macromolecules such as protein.

RESULTS AND DISCUSSION

A series of structurally related derivatives were designed. Those derivatives obeying rule of 5 were selected for docking studies. The structures and molecular descriptors of selected derivatives and standard are depicted in the Table 1. Lipinski's rule of five analysis revealed that all five derivatives were likely to have physical and chemical properties to be orally bioavailable. Docking studies of selected five derivatives (PTC 1, PTC-2, PTC 3, PTC -4 and PTC-5) were carried out using AutoDock Vina with protein ID 5gwk. Schematic 2D representation of docked complex of selected chalcone derivatives and standard (Etoposide) with protein (PDB ID: 5gwk) was visualized using PyMOL. Docking score of derivatives and standard with protein shown in Table 2.

Docking results revealed that compounds PTC-3 and PTC-5 have high binding energy which indicates that these two derivatives possess high affinity and very good interaction within the binding site of 5gwk. The five derivatives and standard exhibited high binding energy due to polar interaction like hydrogen bonding. PTC-1 displays hydrogen bond interaction with ARG-672, MET-672, TYR-757, ARG-673, PTC-2 with ARG-727, GLY-1007, PTC-3 with GLU-839, ARG-727, LEU-722, GLU-712, PTC-4 with GLY-1007, LEU-1003, ARG-727and PTC-5 with ARG-673, GLY-1007. The standard Etoposide displays hydrogen bond interaction with GLY-749, GLU-753, LYS-743, GLY-617, THR-618, LYS-611, GLU-741. Hence these derivatives were expected to have good *in vitro* and *in vivo* anticancer activity.

CONCLUSION

Cancer, a generic term, which is used to refer a group of diseases that can affect any part of the body. It is also known by the name malignant tumor and neoplasm. It involves abnormal cell growth with the potential of spreading to other parts of body known as metastasis, which is the primary cause of death from cancer. Topoisomerase enzyme is one of the effective targets for cancer therapy and development of novel anticancer drug. From the docking studies of derivatives with topoisomerase II, we have found that derivatives PTC-3 and PTC-5 bound to the active pockets of 5gwk with high binding energy and good binding pose. The good binding affinity of the derivatives was due to polar interactions and hydrogen bonding. So, derivatives PTC-3 and PTC-5 are expected to give good *in vitro* and *in vivo* anticancer activity. This may be considered in the design and discovery of ideal Topoisomerase II. Further modification can be carried out to develop better anticancer agents.

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Table1: Molecular Descriptor Analysis Of Selected Derivatives

Compound Code	Structure	M.W	HA	HD	Log P	nrotb	Nviolation
PTC-1		407.49	5	2	4.40	5	0
PTC-2		377.46	4	2	4.38	4	0
PTC-3		379.44	5	4	3.78	3	0
PTC-4		363.44	4	3	4.07	3	0
PTC-5		391.49	4	1	4.69	5	0

| Etoposide | | 588.56 | 13 | 3 | 0.70 | 5 | 2 |

HA: number of hydrogen bond acceptor, HD: number of hydrogen bond donor, nrotb: Number of rotatable bonds
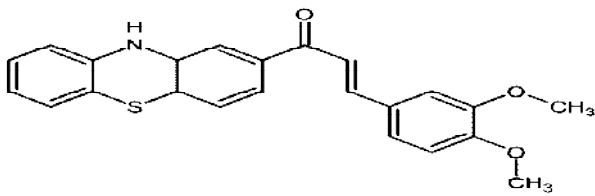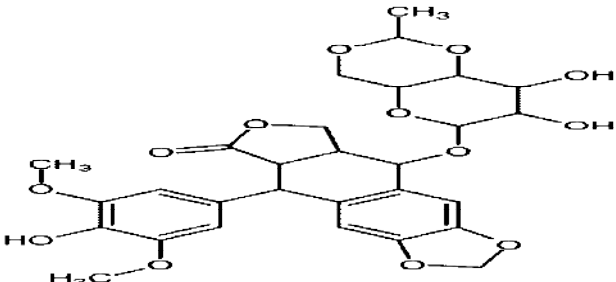
Table 2: Docking Score Of Derivatives With Protein (Pdb Id: 5gwk)

Compound Code	Structure	Docking Score (Kcal/mol)
PTC-1		-9.0
PTC-2		-9.1
PTC-3		-9.4
PTC-4		-9.1

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PTC-5		-9.2
Standard Drug (Etoposide)		-9.7
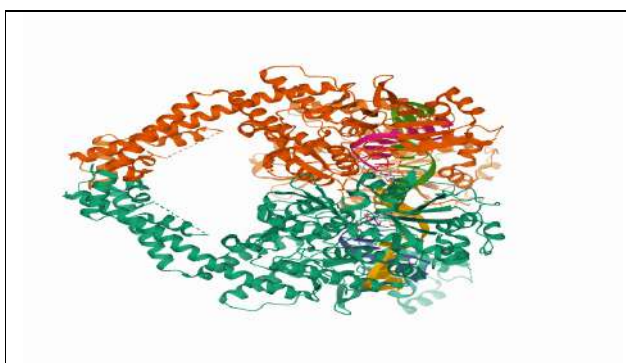
Figure 1: Structure of topoisomerase II
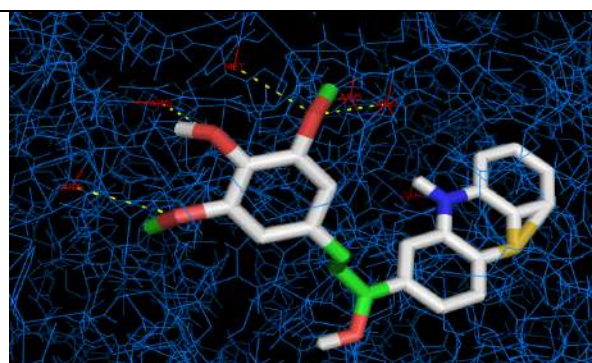
Figure 2: Binding pose ofcompound PTC-1 with 5gwk

Figure 3: Binding pose of compound PTC-2 with 5gwk
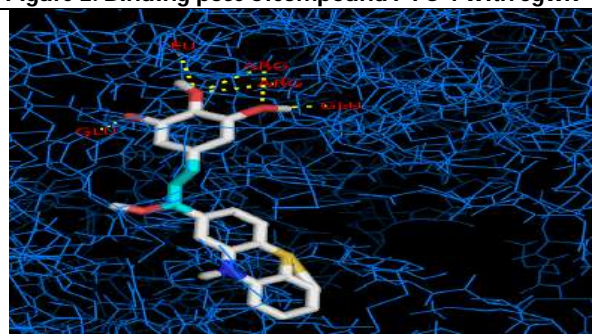
Figure 4: Binding pose of compound PTC-3 with 5gwk

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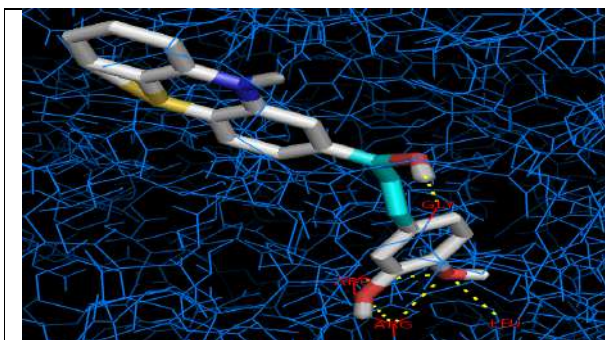
Figure 5: Binding pose of compound PTC-4 with 5gwk
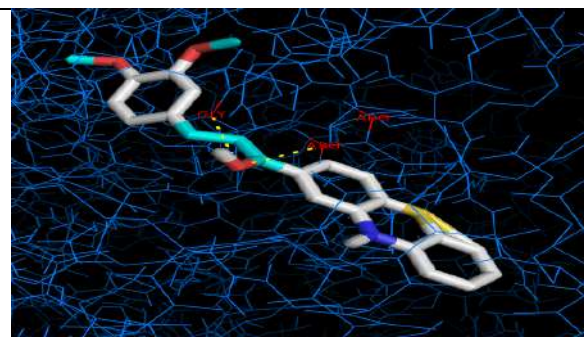
Figure 6: Binding pose of compound PTC-5 with 5gwk
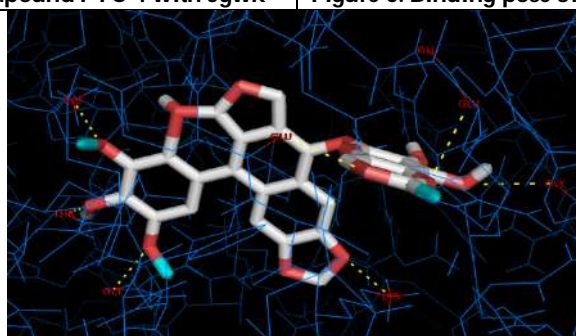
Figure 7: Binding pose of standard drug etoposide with 5gwk

RESEARCH ARTICLE

M/G/1 Queue with Two Types of Service and Multiple Server Vacation

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ABSTRACT

We analyze the steady state behavior of an M/G/1 queue with two types of service subject to multiple server vacation. The server provides two types of service type 1 and type 2 and each arriving customer has the option to choose either type of service. Just before the service of a customer starts, he may choose type 1 service with probability p_1 or type 2 service with probability p_2. If there is no customer waiting in the system, then the server goes for vacation with random duration. On returning from vacation if the server again founds no customer waiting in the system then it goes for vacation again. The server continues to go for vacation until he finds at least one customer in the system. So, the server takes multiple vacation. We obtain time dependent as well as steady state probability generating function for the number in the system. For steady state we obtain explicitly the mean number and the mean waiting time in the system and for the queue. Results for some special cases of interest are derived.

Keywords: Poisson arrivals, Probability generating function, transient state, steady state, supplementary variable technique.

INTRODUCTION

There is natural interest in the study of queueing systems with server vacations or interruptions. Queueing systems that allow servers to take vacation have wide range of applications in many engineering systems such as production, manufacturing, communication networks and telecommunication systems. In fact, Queueing models with server vacations have been efficiently studied by many researchers in the last two decades and successfully applied in various practical problems. An excellent survey on the vacation queueing models have been documented in Kleinrock (1976), Cohen (1982), Lavenberg (1988), Takagi (1991) and several others. There are two basic vacation queueing models namely, multiple vacation queueing model and single vacation queueing model. In multiple vacation queueing models, the server keeps on taking sequential vacations until it finds some customers waiting in a queue at a vacation completion epoch; However, in single vacation queueing models, the server takes exactlyone vacation between two sequential busy periods. These two types of vacation models were first introduced by Levy and Yechiali (1976).

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Many researchers have developed several models involving single vacation policy in vacation queueing systems but only few models have been developed with multiple vacations. A non – Markovian batch arrival, general bulk service single-server queueing system with server breakdown and repair subject to multiple vacation and re-service is studied in Ayyappan, and Karpagam (2016).The probability generating function of the queue size at an arbitrary time and some performance measures of the system are derived for this model. An M/G/1 queue with reneging under extended vacation policy is analyzed in Suthersan and Maragathasundari (2019). The transient analysis of an M/M/1 queue subject to multiple server vacations is investigated in Kaliappan and Kasturi (2014). A single server, infinite buffer, bulk service Poisson queue with single and multiple vacation have been analyzed by Tamrakar and Banerjee (2020). Using bivariate probability generating function (PGF) method, the steady-state joint distributions of the queue content and server content (when server is busy)and joint distribution of the queue content and type of the vacation taken by the server (when server is in vacation) have been obtained. A single server queue with random vacation policy have been studied by Priyanka and Choudhury (2021) in which the server takes the maximum number of random vacations till it finds minimum one message (customer) waiting in a queue at a vacation completion epoch. A finite capacity multi-server Markovianqueueing model with Bernoulli feedback, synchronous multiple vacation and customers impatience (balking and reneging) is investigated in Bouchentouf et al. (2020).

The single server queue with two phases of service with vacations has paid attention recently by several researchers. Presently such type of models has been the subject matter of current research mainly due to its applications in computer and communication systems. The steady state behavior of an M/G/1 queue with two types of general heterogeneous service and optional repeated service subject to server's breakdowns occurring randomly at any instant while serving the customers and delayed repair is studied in Choudhury et al.(2018). An M/G/1 queue with two stage heterogeneous service and deterministic server vacations is analyzed by vanitha (2018). An *M/G/1* retrial queueing system with two phases of service of which the second phase is optional and the server operating under Bernoulli vacation schedule is investigated by Pavai Madheswari et al. (2019). In this paper, the joint generating functions of orbit size and server status are derived using supplementary variable technique. A non-Markovianqueueing model with setup time and Balking is considered by Shyamala and Vijayaraj (2020).In this model, the server provides two stages of service subject to Bernoulli vacation. In extension to this, one of the customers impatience called balking has been incorporated which reflects that a customer may decide to get into the system or not, due to impatience. And also assumed that at the end of a busy period, as soon as a batch of customers arrives, the server does not start giving service, but needs a setup time before actually starting its service of the first customer. An M/G/1 queue with two stage heterogeneous service, optional re - service and server vacation is analyzed in Pazhani Bala Murugan and Kalyanaraman (2020). A single server queueing model with two distinct priorities of customers is studied in Nair et al. (2021). A single server GI/M/1 queue with a limited buffer and an energy saving mechanism based on a single working vacation is investigated in Kobielnik and Kempa (2021).

In the literature, there are several works based on vacation queueing systems subject to two phases of service with single vacation policy and feedback. However, no work has been reported in forming a queueing model with two types of service and multiple server vacation. This motivates us to study an M/G/1 queue with two types of service subject to multiple server vacation. The server provides two types of service type 1 and type 2 and each arriving has the option to choose either type of service. Just before the service of a customer starts, he may choose type 1 service with probability p_1 or type 2 service with probability p_2. If there is no customer waiting in the system, then the server goes for vacation with random duration. On returning from vacation if the server again founds no customer waiting in the system then it goes for vacation again. The server continues to go for vacation until he finds at least one customer in the system. So, the server takes multiple vacation. The rest of the paper is organized as follows. The model under consideration is described in section 2. In Section 3, we have analysed the model by giving the definitions and equations governing the system. Using these equations, the probability generating function of queue length are obtained in section 4. In Section 5, we obtain the steady state solution. Some operating characteristics are obtained in Section 6. In Section 7, we derive mean waiting time. Some particular cases for this model are discussed in Section 8.

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Assumptions Underlying the Model

The following assumptions describe the mathematical model

- Customers arrive at the system one by one in according to a Poisson stream with arrival rate $\lambda(> 0)$.
- The server provides two types of service, type 1 and type 2 with the service times having different general distributions. Let $B_1(v)$ and $b_1(v)$ respectively be the distribution and the density function of the type 1 service.
- The type 2 service times are assumed to be general with the distribution function $B_2(v)$ and the density function $b_2(v)$.
- Just before the service of a customer starts he may choose type 1 service with probability p_1 or type 2 service with probability p_2, where $p_1 + p_2 = 1$.
- Further $\mu_i(x)dx$ is the probability of completion of the ith type service given that elapsed time is x, so that

$$\mu_i(x) = \frac{b_i(x)}{1 - B_i(x)}, i = 1,2 \tag{1}$$

and therefore

$$b_i(v) = \mu_i(v)e^{-\int_0^v \mu_i(x)dx}, i = 1,2. \tag{2}$$

- If there is no customer waiting in the system then the server goes for vacation with random duration. It has general distribution with distribution function $V(x)$ and the probability density function $v(x)$. Also let $E(V^l)$ be the lth finite moment of V where $l \geq 1$. On returning from vacation if the server again founds no customer waiting in the system then it goes for vacation again. The server continues to go for vacation until he finds at least one customer in the system. So, the server takes multiple vacation.
- Let $\gamma(x)dx$ be the conditional probability of completion of the vacation during the time interval $(x, x + dx]$ given that the elapsed vacation time is x so that

$$\gamma(x) = \frac{v(x)}{1 - V(x)} \tag{3}$$

and therefore

$$v(t) = \gamma(t)e^{-\int_0^t \gamma(x)dx}. \tag{4}$$

- The customers are served according to first come first serve rule.
- Various stochastic processes involved in the system are independent of each other.

Equations Governing the System

We define

$P_n^{(1)}(x,t)$ = Probability at time t there are $n(\geq 0)$ customers in the queue excluding one customer in the first type of service and the elapsed service time for this customer is x. Consequently $P_n^{(1)}(t) = \int_0^\infty P_n^{(1)}(x,t)dx$ denotes the probability that at time t there are n customers in the queue excluding the one customer in the first type of service irrespective of the value of x.

$P_n^{(2)}(x,t)$ = Probability at time t there are $n(\geq 0)$ customers in the queue excluding one customer in the second type of service and the elapsed service time for this customer is x. Consequently $P_n^{(2)}(t) = \int_0^\infty P_n^{(2)}(x,t)dx$ denotes the probability that at time t there are n customers in the queue excluding the one customer in the second type of service irrespective of the value of x.

$V_n(x,t)$ = Probability at time t the server is on vacation with the elapsed vacation time x and there are $n(\geq 0)$ customers waiting in the queue for service. Consequently $V_n(t) = \int_0^\infty V_n(x,t)dx$ denotes the probability that at time t there are n customers in the queue and the server is on vacation irrespective of the value of x.

The system has then the following set of differential – difference equations

$$\frac{\partial}{\partial x}P_n^{(1)}(x,t) + \frac{\partial}{\partial t}P_n^{(1)}(x,t) + (\lambda + \mu_1(x))P_n^{(1)}(x,t) = \lambda P_{n-1}^{(1)}(x,t), n = 1,2,\dots \tag{5}$$

$$\frac{\partial}{\partial x}P_0^{(1)}(x,t) + \frac{\partial}{\partial t}P_0^{(1)}(x,t) + \big(\lambda + \mu_1(x)\big)P_0^{(1)}(x,t) = 0, \tag{6}$$

$$\frac{\partial}{\partial x}P_n^{(2)}(x,t) + \frac{\partial}{\partial t}P_n^{(2)}(x,t) + \big(\lambda + \mu_2(x)\big)P_n^{(2)}(x,t) = \lambda P_{n-1}^{(2)}(x,t), n = 1,2,\dots \tag{7}$$

$$\frac{\partial}{\partial x}P_0^{(2)}(x,t) + \frac{\partial}{\partial t}P_0^{(2)}(x,t) + \big(\lambda + \mu_2(x)\big)P_0^{(2)}(x,t) = 0, \tag{8}$$

$$\frac{\partial}{\partial x}V_n(x,t) + \frac{\partial}{\partial t}V_n(x,t) + \big(\lambda + \gamma(x)\big)V_n(x,t) = \lambda V_{n-1}(x,t), n = 1,2,\dots \tag{9}$$

$$\frac{\partial}{\partial x}V_0(x,t) + \frac{\partial}{\partial t}V_0(x,t) + \big(\lambda + \gamma(x)\big)V_0(x,t) = 0. \tag{10}$$

Equations (5) – (10) are to be solved subject to the following boundary conditions:

$$P_0^{(1)}(x,t) = p_1\int_0^\infty P_1^{(1)}(x,t)\mu_1(x)dx + p_1\int_0^\infty P_1^{(2)}(x,t)\mu_2(x)dx + p_1\int_0^\infty V_1(x,t)\gamma(x)dx, \tag{11}$$

$$P_n^{(1)}(x,t) = p_1\int_0^\infty P_{n+1}^{(1)}(x,t)\mu_1(x)dx + p_1\int_0^\infty P_{n+1}^{(2)}(x,t)\mu_2(x)dx + p_1\int_0^\infty V_{n+1}(x,t)\gamma(x)dx,$$
$$n = 1,2,\dots, \tag{12}$$

$$P_0^{(2)}(x,t) = p_2\int_0^\infty P_1^{(1)}(x,t)\mu_1(x)dx + p_2\int_0^\infty P_1^{(2)}(x,t)\mu_2(x)dx + p_2\int_0^\infty V_1(x,t)\gamma(x)dx \tag{13}$$

$$P_n^{(2)}(x,t) = p_2\int_0^\infty P_{n+1}^{(1)}(x,t)\mu_1(x)dx + p_2\int_0^\infty P_{n+1}^{(2)}(x,t)\mu_2(x)dx + p_2\int_0^\infty V_{n+1}(x,t)\gamma(x)dx,$$
$$n = 1,2,\dots, \tag{14}$$

$$V_0(x,t) = \int_0^\infty P_0^{(1)}(x,t)\mu_1(x)dx + \int_0^\infty P_0^{(2)}(x,t)\mu_2(x)dx + \int_0^\infty V_0(x,t)\gamma(x)dx, \tag{15}$$

$$V_n(x,t) = 0, n = 1,2,\dots. \tag{16}$$

We assume that initially there is no customer in the system and the server is not under vacation. So the initial conditions are

$$V_0(0) = V_n(0) = 0 \text{ and } P_n^{(j)}(0) = 0 \text{ for } n = 0,1,2,\dots, j = 1,2. \tag{17}$$

Generating Functions of the Queue Length: The Time - Dependent Solution

In this section we obtain the transient solution of the model described by the above set of differential – difference equations.

We define the probability generating functions

$$\left.\begin{array}{l} P^{(1)}(x,z,t) = \sum_{n=0}^\infty z^n P_n^{(1)}(x,t), P^{(1)}(z,t) = \sum_{n=0}^\infty z^n P_n^{(1)}(t), \\ P^{(2)}(x,z,t) = \sum_{n=0}^\infty z^n P_n^{(2)}(x,t), P^{(2)}(z,t) = \sum_{n=0}^\infty z^n P_n^{(2)}(t) \\ V(x,z,t) = \sum_{n=0}^\infty z^n V_n(x,t), V(z,t) = \sum_{n=0}^\infty z^n V_n(t) \end{array}\right\} \tag{18}$$

which are convergent inside the circle given by $|z| \le 1$ and define the Laplace transform of a function $f(t)$ as

$$\overline{f(s)} = \int_0^\infty e^{-st}f(t)dt, \Re(s) > 0. \tag{19}$$

Taking Laplace transforms of equations (5) - (16) and using (17) we obtain

$$\frac{\partial}{\partial x}\overline{P_n}^{(1)}(x,s) + \big(s + \lambda + \mu_1(x)\big)\overline{P_n}^{(1)}(x,s) = \lambda\overline{P_{n-1}}^{(1)}(x,s), n = 1,2,\dots, \tag{20}$$

$$\frac{\partial}{\partial x}\overline{P_0}^{(1)}(x,s) + \big(s + \lambda + \mu_1(x)\big)\overline{P_0}^{(1)}(x,s) = 0, \tag{21}$$

$$\frac{\partial}{\partial x}\overline{P_n}^{(2)}(x,s) + \big(s + \lambda + \mu_2(x)\big)\overline{P_n}^{(2)}(x,s) = \lambda\overline{P_{n-1}}^{(2)}(x,s), n = 1,2,\dots, \tag{22}$$

$$\frac{\partial}{\partial x}\overline{P_0}^{(2)}(x,s) + \big(s + \lambda + \mu_2(x)\big)\overline{P_0}^{(2)}(x,s) = 0, \tag{23}$$

$$\frac{\partial}{\partial x}\overline{V_n}(x,s) + \big(s + \lambda + \gamma(x)\big)\overline{V_n}(x,s) = \lambda\overline{V_{n-1}}(x,s), n = 1,2,\dots \tag{24}$$

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$$\frac{\partial}{\partial x}\overline{V}_0(x,s) + \big(s + \lambda + \gamma(x)\big)\overline{V}_0(x,s) = 0, \tag{25}$$

$$\overline{P}_0^{(1)}(0,s) = p_1 \int_0^\infty \overline{P}_1^{(1)}(x,s)\mu_1(x)dx + p_1 \int_0^\infty \overline{P}_1^{(2)}(x,s)\mu_2(x)dx + p_1 \int_0^\infty \overline{V}_1(x,s)\gamma(x)dx, \tag{26}$$

$$\overline{P}_n^{(1)}(0,s) = p_1 \int_0^\infty \overline{P}_{n+1}^{(1)}(x,s)\mu_1(x)dx + p_1 \int_0^\infty \overline{P}_{n+1}^{(2)}(x,s)\mu_2(x)dx$$

$$+p_1 \int_0^\infty \overline{V}_{n+1}(x,s)\gamma(x)dx, n = 1,2,\dots \tag{27}$$

$$\overline{P}_0^{(2)}(0,s) = p_2 \int_0^\infty \overline{P}_1^{(1)}(x,s)\mu_1(x)dx + p_2 \int_0^\infty \overline{P}_1^{(2)}(x,s)\mu_2(x)dx + p_2 \int_0^\infty \overline{V}_1(x,s)\gamma(x)dx, \tag{28}$$

$$\overline{P}_n^{(2)}(0,s) = p_2 \int_0^\infty \overline{P}_{n+1}^{(1)}(x,s)\mu_1(x)dx + p_2 \int_0^\infty \overline{P}_{n+1}^{(2)}(x,s)\mu_2(x)dx$$

$$+p_2 \int_0^\infty \overline{V}_{n+1}(x,s)\gamma(x)dx, n = 1,2,\dots \tag{29}$$

$$\overline{V}_0(0,s) = \int_0^\infty \overline{P}_0^{(1)}(x,s)\mu_1(x)dx + \int_0^\infty \overline{P}_0^{(2)}(x,s)\mu_2(x)dx + \int_0^\infty \overline{V}_0(x,s)\gamma(x)dx, \tag{30}$$

$$\overline{V}_n(0,s) = 0, n = 1,2,\dots . \tag{31}$$

We multiply both sides of equations (20) and (21) by suitable powers of z, sum over n and use equation (18) and simplify

$$\frac{\partial}{\partial x}\overline{P}^{(1)}(x,z,s) + \big(s + \lambda - \lambda z + \mu_1(x)\big)\overline{P}^{(1)}(x,z,s) = 0, \tag{32}$$

Performing similar operations on equations (22) to (25) we obtain,

$$\frac{\partial}{\partial x}\overline{P}^{(2)}(x,z,s) + \big(s + \lambda - \lambda z + \mu_2(x)\big)\overline{P}^{(2)}(x,z,s) = 0, \tag{33}$$

$$\frac{\partial}{\partial x}\overline{V}(x,z,s) + (s + \lambda - \lambda z + \gamma(x))\overline{V}(x,z,s) = 0. \tag{34}$$

For the boundary conditions we multiply both sides of equation (26) by z, multiply both sides of equation (27) by z^{n+1}, sum over n from 1 to ∞, add the two results and use equation (18) to get

$$z\,\overline{P}^{(1)}(0,z,s) = p_1 \left[\begin{matrix} \int_0^\infty \overline{V}(x,z,s)\gamma(x)dx + \int_0^\infty \overline{P}^{(1)}(x,z,s)\mu_1(x)dx \\ + \int_0^\infty \overline{P}^{(2)}(x,z,s)\mu_2(x)dx \end{matrix} \right]$$

$$-p_1 \left[\int_0^\infty \overline{V}_0(x,s)\gamma(x)dx + \int_0^\infty \overline{P}_0^{(1)}(x,s)\mu_1(x)dx + \int_0^\infty \overline{P}_0^{(2)}(x,s)\mu_2(x)dx \right]. \tag{35}$$

Perform similar to equations (28) to (29) and (30) to (31), we obtain

$$z\,\overline{P}^{(2)}(0,z,s) = p_2 \left[\begin{matrix} \int_0^\infty \overline{V}(x,z,s)\gamma(x)dx + \int_0^\infty \overline{P}^{(1)}(x,z,s)\mu_1(x)dx \\ + \int_0^\infty \overline{P}^{(2)}(x,z,s)\mu_2(x)dx \end{matrix} \right]$$

$$-p_2 \left[\int_0^\infty \overline{V}_0(x,s)\gamma(x)dx + \int_0^\infty \overline{P}_0^{(1)}(x,s)\mu_1(x)dx + \int_0^\infty \overline{P}_0^{(2)}(x,s)\mu_2(x)dx \right], \tag{36}$$

$$\overline{V}(0,z,s) = \overline{V}_0(0,s) . \tag{37}$$

Using equation (30), equations (35) and (36) become

$$z\,\overline{P}^{(1)}(0,z,s) = p_1 \left[\begin{matrix} \int_0^\infty \overline{V}(x,z,s)\gamma(x)dx + \int_0^\infty \overline{P}^{(1)}(x,z,s)\mu_1(x)dx \\ + \int_0^\infty \overline{P}^{(2)}(x,z,s)\mu_2(x)dx \end{matrix} \right] - p_1\overline{V}_0(0,s), \tag{38}$$

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$$z\,\overline{P}^{(2)}(0,z,s) = p_2\left[\int_0^\infty \overline{V}(x,z,s)\gamma(x)dx + \int_0^\infty \overline{P}^{(1)}(x,z,s)\mu_1(x)dx + \int_0^\infty \overline{P}^{(2)}(x,z,s)\mu_2(x)dx\right] - p_2\overline{V_0}(0,s). \tag{39}$$

Integrating equations (32), (33) and (34) between 0 to x yield

$$\overline{P}^{(1)}(x,z,s) = \overline{P}^{(1)}(0,z,s)e^{-(s+\lambda-\lambda z)x-\int_0^\infty \mu_1(t)dt}, \tag{40}$$

$$\overline{P}^{(2)}(x,z,s) = \overline{P}^{(2)}(0,z,s)e^{-(s+\lambda-\lambda z)x-\int_0^\infty \mu_2(t)dt}, \tag{41}$$

$$\overline{V}(x,z,s) = \overline{V}(0,z,s)e^{-(s+\lambda-\lambda z)x-\int_0^\infty \gamma(t)dt}, \tag{42}$$

where $\overline{P}^{(1)}(0,z,s), \overline{P}^{(2)}(0,z,s)$ and $\overline{V}(0,z,s)$ are given by equations (38), (39) and (37) .

Again integrating equation (40) with respect to x exhibits

$$\overline{P}^{(1)}(z,s) = \overline{P}^{(1)}(0,z,s)\left[\frac{1-\overline{B_1}(s+\lambda-\lambda z)}{s+\lambda-\lambda z}\right] \tag{43}$$

where

$$\overline{B_!}(s+\lambda-\lambda z) = \int_0^\infty e^{-(s+\lambda-\lambda z)x}\,\mathrm{dB_1(x)} \tag{44}$$

is the Laplace - Steiltjes transform of the type 1 service time.

Now multiplying both sides of equation of (40) by $\mu_1(x)$ and integrating over x, we obtain,

$$\int_0^\infty \overline{P}^{(1)}(x,z,s)\mu_1(x)dx = \overline{P}^{(1)}(0,z,s)\overline{B_1}(s+\lambda-\lambda z). \tag{45}$$

We now integrate equation (33) with respect to x to get

$$\overline{P}^{(2)}(z,s) = \overline{P}^{(2)}(0,z,s)\left[\frac{1-\overline{B_2}(s+\lambda-\lambda z)}{s+\lambda-\lambda z}\right] \tag{46}$$

where

$$\overline{B_2}(s+\lambda-\lambda z) = \int_0^\infty e^{-(s+\lambda-\lambda z)x}\,\mathrm{dB_2(x)} \tag{47}$$

is the Laplace - Steiltjes transform of the type 2 service time.

We see that by virtue of equation (41), we get

$$\int_0^\infty \overline{P}^{(2)}(x,z,s)\mu_2(x)dx = \overline{P}^{(2)}(0,z,s)\overline{B_2}(s+\lambda-\lambda z) . \tag{48}$$

Perform similar operations on equation (42), we obtain

$$\overline{V}(z,s) = \overline{V}(0,z,s)\left[\frac{1-\overline{V}(s+\lambda-\lambda z)}{s+\lambda-\lambda z}\right], \tag{49}$$

where

$$\overline{V}(s+\lambda-\lambda z) = \int_0^\infty e^{-(s+\lambda-\lambda z)x}\,\mathrm{dV(x)} \tag{50}$$

is the Laplace - Steiltjes transform of the vacation time.

We see that by virtue of equation (42), we have

$$\int_0^\infty \overline{V}(x,z,s)\gamma(x)dx = \overline{V}(0,z,s)\overline{V}(s+\lambda-\lambda z). \tag{51}$$

We now substitute the value of $\int_0^\infty \overline{V}(x,z,s)\gamma(x)dx$ from equation (51) into equations (38) and (39) and also making use of equations (37), (45) and (48), we obtain after simplifications

$$z\,\overline{P}^{(1)}(0,z,s) = p_1\overline{V}(0,s)\left[\overline{V}(s+\lambda-\lambda z)-1\right] + p_1\overline{P}^{(1)}(0,z,s)\overline{B_1}(s+\lambda-\lambda z)$$

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$$+ p_1 \overline{P}^{(2)}(0,z,s)\overline{B_2}(s + \lambda - \lambda z) , \tag{52}$$

$$z\,\overline{P}^{(2)}(0,z,s) = p_2\overline{V}(0,s)\big[\overline{V}(s + \lambda - \lambda z) - 1\big] + p_2\overline{P}^{(1)}(0,z,s)\overline{B_1}(s + \lambda - \lambda z)$$

$$+ p_2\overline{P}^{(2)}(0,z,s)\overline{B_2}(s + \lambda - \lambda z) . \tag{53}$$

Now we write equations (52) and (53) in matrix form as

$$\begin{vmatrix} h_1(z) & -k_2(z) \\ -k_1(z) & h_2(z) \end{vmatrix} \begin{vmatrix} \overline{P}^{(1)}(0,z,s) \\ \overline{P}^{(2)}(0,z,s) \end{vmatrix} = \begin{vmatrix} p_1\overline{V_0}(0,s)\big[\overline{V}(s + \lambda - \lambda z) - 1\big] \\ p_2\overline{V_0}(0,s)\big[\overline{V}(s + \lambda - \lambda z) - 1\big] \end{vmatrix} , \tag{54}$$

where

$$h_1(z) = z - p_1\overline{B_1}(s + \lambda - \lambda z) , \tag{55}$$

$$h_2(z) = z - p_2\overline{B_2}(s + \lambda - \lambda z) , \tag{56}$$

$$k_1(z) = p_2\overline{B_1}(s + \lambda - \lambda z) , \tag{57}$$

and

$$k_2(z) = p_1\overline{B_2}(s + \lambda - \lambda z) . \tag{58}$$

We solve the system (54) simultaneously for $\overline{P}^{(1)}(0,z,s)$ and $\overline{P}^{(2)}(0,z,s)$ and obtain

$$\overline{P}^{(1)}(0,z,s) = \frac{\begin{vmatrix} p_1\overline{V_0}(0,s)[\overline{V}(s+\lambda-\lambda z)-1] & -k_2(z) \\ p_2\overline{V_0}(0,s)[\overline{V}(s+\lambda-\lambda z)-1] & h_2(z) \end{vmatrix}}{\begin{vmatrix} h_1(z) & -k_2(z) \\ -k_1(z) & h_2(z) \end{vmatrix}} ,$$

$$= \frac{\overline{V_0}(0,s)[\overline{V}(s+\lambda-\lambda z)-1][p_1 h_2(z)+p_2 k_2(z)]}{h_1(z)h_2(z)-k_1(z)k_2(z)} \tag{59}$$

and

$$\overline{P}^{(2)}(0,z,s) = \frac{\begin{vmatrix} h_1(z) & p_1\overline{V_0}(0,s)[\overline{V}(s + \lambda - \lambda z) - 1] \\ -k_1(z) & p_2\overline{V_0}(0,s)[\overline{V}(s + \lambda - \lambda z) - 1] \end{vmatrix}}{\begin{vmatrix} h_1(z) & -k_2(z \\ -k_1(z) & h_2(z) \end{vmatrix}}$$

$$= \frac{\overline{V_0}(0,s)[\overline{V}(s+\lambda-\lambda z)-1][p_2 h_1(z)+p_1 k_1(z)]}{h_1(z)h_2(z)-k_1(z)k_2(z)} . \tag{60}$$

Using equation (37) in equation (49), we yield

$$\overline{V}(z,s) = \overline{V_0}(0,s)\left[\frac{1-\overline{V}(s+\lambda-\lambda z)}{(s+\lambda-\lambda z)}\right] . \tag{61}$$

Now using equation (59) in equation (43), we get

$$\overline{P}^{(1)}(z,s) = \left[\frac{\overline{V_0}(0,s)[\overline{V}(s+\lambda-\lambda z)-1][p_1 h_2(z)+p_2 k_2(z)]}{h_1(z)h_2(z)-k_1(z)k_2(z)}\right]\left[\frac{1-\overline{B_1}(s+\lambda-\lambda z)}{(s+\lambda-\lambda z)}\right] . \tag{62}$$

Also from equations (46) and (60), we obtain

$$\overline{P}^{(2)}(z,s) = \left[\frac{\overline{V_0}(0,s)[\overline{V}(s+\lambda-\lambda z)-1][p_2 h_1(z)+p_1 k_1(z)]}{h_1(z)h_2(z)-k_1(z)k_2(z)}\right]\left[\frac{1-\overline{B_2}(s+\lambda-\lambda z)}{(s+\lambda-\lambda z)}\right]. \tag{63}$$

where $h_1(z), h_2(z), k_1(z)$ and $k_2(z)$ are given by equations (55) to (58) .

Thus $\overline{P}^{(1)}(z,s)$, $\overline{P}^{(2)}(z,s)$ and $\overline{V}(z,s)$ can be completely determined from equations (62), (63) and (61) respectively.

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THE STEADY STATE RESULTS

In this section, we shall derive the steady state probability distribution for our queueing model. To define the steady state probabilities, we suppress the argument t wherever it appears in the time-dependent analysis. This can be obtained by applying the well-known Tauberian property,

$$\lim_{n \to 0} s\,\overline{f}(s) = \lim_{t \to \infty} f(t).$$

In order to determine $\overline{P}^{(1)}(z,s), \overline{P}^{(2)}(z,s)$ and $\overline{V}(z,s)$ completely, we have yet to determine the only unknown $V_0(0)$ which appear in the numerators of equations (62), (63) and (61) respectively. For that purpose, we shall use the normalizing condition

$$P^{(1)}(1) + P^{(2)}(1) + V(1) = 1 . \tag{64}$$

Thus multiplying both sides of equations (62), (63) and (61) by s, taking limit as $s \to 0$, applying property (64) and simplifying we have

$$P^{(1)}(z) = \left[\overline{V}(\lambda - \lambda z) - 1\right]\left[\frac{p_1 h_2(z) + p_2 k_2(z)}{h_1(z)h_2(z) - k_1(z)k_2(z)}\right] V_0(0) \left[\frac{1 - \overline{B_1}(\lambda - \lambda z)}{(\lambda - \lambda z)}\right], \tag{65}$$

$$P^{(2)}(z) = \left[\overline{V}(\lambda - \lambda z) - 1\right]\left[\frac{p_2 h_1(z) + p_1 k_1(z)}{h_1(z)h_2(z) - k_1(z)k_2(z)}\right] V_0(0) \left[\frac{1 - \overline{B_2}(\lambda - \lambda z)}{(\lambda - \lambda z)}\right] \tag{66}$$

and $V(z) = \left[\frac{1 - \overline{V}(\lambda - \lambda z)}{\lambda - \lambda z}\right] V_0(0). \tag{67}$

We see that for $z = 1$, $P^{(1)}(z), P^{(2)}(z)$ and V(z) in equations (65) - (67) are indeterminate of the $\frac{0}{0}$ form. Therefore, we apply L'Hopital's rule on equations (65) - (67) using the fact that $\overline{B_1}(0) = 1, -\overline{B_1}'(0) = \frac{1}{\mu_1}, \overline{B_2}(0) = 1, -\overline{B_2}'(0) = \frac{1}{\mu_2}, \overline{V}(0) = 1, -\overline{V}'(0) = \frac{1}{\gamma} = E(v)$. Thus on simplifying we have

$$P^{(1)}(1) = \left[\frac{p_1 \lambda E(v)E(v_1)}{1 - p_1 \lambda E(v_1) - p_2 \lambda E(v_2)}\right] V_0(0), \tag{68}$$

$$P^{(2)}(1) = \left[\frac{p_2 \lambda E(v)E(v_2)}{1 - p_1 \lambda E(v_1) - p_2 \lambda E(v_2)}\right] V_0(0), \tag{69}$$

and

$$V(1) = V_0(0)E(v) , \tag{70}$$

where $E(v_1), E(v_2)$ and $E(v)$ denote the mean service time of type 1 service, type 2 service and mean vacation time respectively. Note that the results (68) to (70) give the steady state probabilities that the server is providing type 1 service, type 2 service and under vacation respectively.

Now using equations (68) to (70) into the normalizing condition (64) and simplifying we obtain

$$V_0(0) = \left[\frac{1 - p_1 \lambda E(v_1) - p_2 \lambda E(v_2)}{E(v)}\right] . \tag{71}$$

Also from equation (70), we obtain system's utilization factor

$$\rho = [p_1 \lambda E(v_1) + p_2 \lambda E(v_2)] , \tag{72}$$

where $\rho < 1$ is the stability condition under which the steady state exists.

Thus we have now explicitly determined all the steady state probability generating functions $P^{(1)}(z), P^{(2)}(z)$ and $V(z)$ of the queue size.

The Mean Number in the System

Now we define $P(z)$ as the probability generating function of the queue size. Then we have

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$$P(z) = V(z) + z\left(P^{(1)}(z) + P^{(2)}(z)\right). \tag{73}$$

Let L_q and L denote the steady state average queue size and system size respectively. We have $L_q = \frac{d}{dz}P(z)$ at $z = 1$. However since $P(z) = \frac{0}{0}$ at $z = 1$. We use the well-known result in Queueing theory (Kashyap and Chaudhry (1988)) to get

$$L_q = \lim_{z \to 1}\frac{d}{dz}P(z) = P^{'}(1) = \lim_{z \to 1}\frac{D^{'}(z)N^{''}(z) - N^{'}(z)D^{''}(z)}{2\left(D^{'}(z)\right)^2}\left[\frac{V_0(0)}{\lambda}\right],$$

$$= \lim_{z \to 1}\frac{D^{'}(1)N^{''}(1) - N^{'}(1)D^{''}(1)}{2\left(D^{'}(1)\right)^2}\left[\frac{V_0(0)}{\lambda}\right], \tag{74}$$

where primes and double primes in equation (74) denote the first and second derivative at $z = 1$. Using equations (65) to (67) into equation (73), we have

$$P(z) = \frac{N(z)}{D(z)}\left[\frac{V_0(0)}{\lambda}\right] \tag{75}$$

where

$$N(z) = \left[\overline{V}(\lambda - \lambda z) - 1\right]\left[zp_1\overline{B_1}(\lambda - \lambda z) + zp_2\overline{B_2}(\lambda - \lambda z)\right] \tag{76}$$

and

$$D(z) = \left[z - p_1\overline{B_1}(\lambda - \lambda z)\right]\left[z - p_1\overline{B_1}(\lambda - \lambda z)\right] - p_1p_2\overline{B_1}(\lambda - \lambda z)\overline{B_2}(\lambda - \lambda z). \tag{77}$$

Carrying out the derivatives at $= 1$, we have

$$N^{'}(1) = \lambda E(v), \tag{78}$$

$$N^{''}(1) = \lambda^2 E(v^2) + 2\lambda E(v) + 2\lambda^2 p_1 E(v)E(v_1) + 2\lambda^2 p_2 E(v)E(v_2), \tag{79}$$

$$D^{'}(1) = 1 - p_1\lambda E(v_1) - p_2\lambda E(v_2), \tag{80}$$

$$D^{''}(1) = -\lambda^2[p_1 E(v_1^2) + p_2 E(v_2^2)] + 2[1 - p_1\lambda E(v_1) - p_2\lambda E(v_2)]. \tag{81}$$

where $E(v_1^2), E(v_2^2)$ and $E(v^2)$ denote the second moments of the type 1 service, type 2 service and vacation time respectively. Using equations (78) to (81) into equation (74), we have obtained L_q in closed form. Further we find the average system size L using Little's formula. Thus we have

$$L = L_q + \rho \tag{82}$$

where L_q has been found in equation (74) and ρ is obtained from equation (72).

The Mean Waiting Time

Let W_q and W denote the mean waiting time in the queue and the system respectively. Then using Little's formula we obtain

$$W_q = \frac{L_q}{\lambda} \tag{83}$$

$$W = \frac{L}{\lambda} \tag{84}$$

where L_q and L have been found in equations (74) and (82).

Special Cases

Case 1: First stage of service follows exponential distribution

In this case, we put $\overline{B_1}(\lambda - \lambda z) = \frac{\mu_1}{\lambda - \lambda z + \mu_1}, E(v_1) = \frac{1}{\mu_1}, E(v_1^2) = \frac{2}{\mu_1^2}$ in equation (75) so that

$$P(z) = \frac{\left[\overline{V}(\lambda - \lambda z) - 1\right]\left[zp_1\frac{\mu_1}{\lambda - \lambda z + \mu_1} + zp_2\overline{B_2}(\lambda - \lambda z)\right]}{z^2 - zp_1\frac{\mu_1}{\lambda - \lambda z + \mu_1} - zp_2\overline{B_2}(\lambda - \lambda z)}\left[\frac{V_0(0)}{\lambda}\right]. \tag{85}$$

Further we have $V_0(0) = \left[\frac{1 - p_1\frac{\lambda}{\mu_1} - p_2\lambda E(v_2)}{E(v)}\right], \tag{86}$

$$\rho = \left[p_1\frac{\lambda}{\mu_1} + p_2\lambda E(v_2)\right] < 1 \tag{87}$$

and L_q is given by (74), where

$$N^{'}(1) = \lambda E(v), \tag{88}$$

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$$N^{''}(1) = \lambda^2 \frac{2}{\mu_1^2} + 2\lambda E(v) + 2\lambda^2 p_1 E(v)\frac{1}{\mu_1} + 2\lambda^2 p_2 E(v)E(v_2), \tag{89}$$

$$D^{'}(1) = 1 - p_1\frac{\lambda}{\mu_1} - p_2 \lambda E(v_2), \tag{90}$$

$$D^{''}(1) = -\lambda^2\left[p_1\frac{2}{\mu_1^2} + p_2 E(v_2^2)\right] + 2\left[1 - p_1\frac{\lambda}{\mu_1} - p_2 \lambda E(v_2)\right]. \tag{91}$$

In addition L, W and W_q for this case can also be found from the main results.

Case 2: Both services follow exponential distribution

In this case, we let $\overline{B_2}(\lambda - \lambda z) = \frac{\mu_2}{\lambda - \lambda z + \mu_2}, E(v_2) = \frac{1}{\mu_2}, E(v_2^2) = \frac{2}{\mu_2^2}$ in all the results obtained in Case 1 so that

$$P(z) = \frac{[\overline{V}(\lambda - \lambda z) - 1]\left[zp_1\frac{\mu_1}{\lambda - \lambda z + \mu_1} + zp_2\frac{\mu_2}{\lambda - \lambda z + \mu_2}\right]}{z^2 - zp_1\frac{\mu_1}{\lambda - \lambda z + \mu_1} - zp_2\frac{\mu_2}{\lambda - \lambda z + \mu_2}}\left[\frac{V_0(0)}{\lambda}\right], \tag{92}$$

$$V_0(0) = \left[\frac{1 - p_1\frac{\lambda}{\mu_1} - p_2\frac{\lambda}{\mu_2}}{E(v)}\right], \tag{93}$$

$$\rho = \left[p_1\frac{\lambda}{\mu_1} + p_2\frac{\lambda}{\mu_2}\right] < 1 \tag{94}$$

and L_q is given by (74), where

$$N^{'}(1) = \lambda E(v), \tag{95}$$

$$N^{''}(1) = \lambda^2 \frac{2}{\mu_1^2} + 2\lambda E(v) + 2\lambda^2 p_1 E(v)\frac{1}{\mu_1} + 2\lambda^2 p_2 E(v)\frac{1}{\mu_2}, \tag{96}$$

$$D^{'}(1) = 1 - p_1\frac{\lambda}{\mu_1} - p_2\frac{\lambda}{\mu_2}, \tag{97}$$

$$D^{''}(1) = -\lambda^2\left[p_1\frac{2}{\mu_1^2} + p_2\frac{2}{\mu_2^2}\right] + 2\left[1 - p_1\frac{\lambda}{\mu_1} - p_2\frac{\lambda}{\mu_2}\right]. \tag{98}$$

In addition L, W and W_q for this case can also be found from the main results.

Case 3: Vacation period follows exponential distribution

In this case, we let $\overline{V}(\lambda - \lambda z) = \frac{\gamma}{\lambda - \lambda z + \gamma}, E(v) = \frac{1}{\gamma}, E(v^2) = \frac{2}{\gamma^2}$ in all the results obtained in Case 2.

Case 4: No server vacation

In this case, we let $\overline{V}(\lambda - \lambda z) = z$ in equation (76) so that

$$P(z) = \frac{N(z)}{D(z)}\left[\frac{V_0(0)}{\lambda}\right],$$

where

$$N(z) = [z - 1]\left[zp_1\overline{B_1}(\lambda - \lambda z) + zp_2\overline{B_2}(\lambda - \lambda z)\right] \tag{99}$$

and $D(z)$ is given by equation (77).

Further L_q, L, W_q and W for this case can be derived from the main results.

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RESEARCH ARTICLE

Recent Approaches to Nasal Drug Delivery: Mucoadhesive Polymers

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ABSTRACT

About 90% of drugs are delivered orally (e.g., tablets, capsules) or by parenteral injection in the case of chemicals that are degraded in the gastrointestinal environment or are not absorbed. As a result, an easy-to-use alternative nasal drug delivery system was created. Nasal drug delivery methods can be used locally as well as systemically. The blood–brain barrier (BBB) is a barrier that prevents therapeutic drugs from reaching the brain. Polymers are the foundation of a pharmaceutical medication delivery system because they regulate the drug's release from the device. The mucoadhesion process utilizing a polymeric drug delivery platform includes actions such as wetting, adsorption, and interpenetration of polymer chains. As a result, APIs have been delivered to a variety of mucosal-enveloped organelles for local or systemic activity via mucoadhesive techniques. The nasal mucosa could be a useful way for delivering drugs in the body. The nasal route avoids first-pass hepatic metabolism, which is one of its most important properties. The future potential of research for practical applications have forced the field's development.

INTRODUCTION

Past few decades, Nasal medication delivery has shifted its focus in recent decades to local and systemic therapeutic delivery. It is mostly due to the patient's comfort and compliance, as well as reliable medication administration, not just for local medicines but also for systemic activities [1]. The nasal drug delivery method, on the other hand, has been shown to be a potential route for systemic treatment for medicines and biomolecules that are prone to enzymic degradation and hepatic first pass metabolism [2]. The binding of a polymers to a biological substrate is known as bioadhesion. Mucoadhesion method has been widely used as a way of getting target drug delivery in pharmaceutical

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developments by mixing polymers of hydrophilic mucoadhesion with Active Pharmaceutical Ingredients (API) [3]. Nasal administration is an intriguing alternative to parenteral administration, which are uncomfortable, and oral delivery, which can lead to excessively poor bioavailability. The nasal epithelium is extremely permeable, the submucosa is well-vascularized, and first-pass metabolism is omitted after nasal delivery. The nasal cavity's relatively large surface area and relatively high blood flow, which facilitates fast absorption, are two further appealing qualities. However, there are a substantial number of naturally occurring biodegradable polymers used in pharmaceutical, medical, and biological applications [4,5]. Polymeric characteristics such as degree of crosslinking, length of the chain, and different polymeric functional groups which influence mucoadhesion. Mucoadhesive methods are commonly utilized to transfer active chemicals to mucosal-covered organelles for local or systemic impact [6]. This chapter briefly reviews the history, categorization, and kinds of biodegradable polymers, biodegradation methods, biodegradation variables, medical, biomedical, and pharmaceutical uses of diverse biopolymers, and their current and future commercial and regulatory scenarios, mucoadhesive techniques are routinely used.

Origin

The nasal mucosa has long been thought to be a promising route for delivering systemic drugs. The size of normal human nasal mucosa is 150 cm and it is covered by a thick vascular network, for good absorption. The nasal epithelium has a good permeability, [7] Thus the nasal lumen is separated from the vast vasculature within the lamina propria by only two cells' layers. The nasal cavity is an appealing route for medication delivery because of these properties, but they also make nasal mucosa cells are risky to the deleterious effects of intranasally delivered drugs and excipients. The nasal cavity gives a wide, better vascularized surface area via first-pass metabolism are omitted because blood is delivered directly into the systemic circulation from the nose. This is one of the major benefits of intranasal medication administration. Nasal delivery has been achieved using a variety of methods, including solutions, powders, gels, and microparticles. The highly often used intranasal APIs for quick alleviation of nasal congestion are solutions containing sympathomimetic vasoconstrictors. Because the nose is where vasoconstriction happens the most, individuals with high blood pressure can benefit from local administration of these alpha-adrenergic stimulators. Intranasal medication delivery has been used to create a distant systemic effect in addition to local effects [8].

Mucoadhesive mechanism

Mucoadhesion is caused by the interaction of the mucoadhesive polymer with the mucus produced by the submucosal glands after nasal administration. The enlarged mucoadhesive polymer subsequently enters the tissue crevices, where it interacts with the mucus protein chains. When the polymer–water contact exceeds the polymer–polymer interaction, sufficient free polymer chains will be accessible for polymer-biological tissue interaction. For optimal mucoadhesion, a certain level of hydration is essential. The mechanism of mucoadhesion may be classified into different parts.

Contact stage: The first wetting that happens between the adhesive and the membrane is known as the contact stage. This can happen mechanically by bringing the two surfaces together, or it can happen through the bodily systems, as when particles are inhaled and deposited in the nasal cavity. The DLVO theory may be used to explain the principles of initial adsorption of tiny molecular adsorbates.

Consolation stage: The formation of adhesive contacts to strengthen strong or lasting adherence is the consolation stage of mucoadhesion. When the surface is subjected to considerable dislodging forces, consolidation variables are critical. There are several stage mucoadhesion hypotheses that explain the consolidation stage, the two most common of which are macromolecular interpenetration and dehydration. [9,10]

Mechanisms of intranasal drug administration

The drug is get absorbed when it passes through the mucous layer of nasal cavity. Charged molecules find it more difficult to pass through mucus than uncharged molecules. Paracellular absorption and transcellular absorption have

been identified as the two primary modes of drug absorption via the nasal mucosa. The medicine diffuses slowly and passively via the watery spaces between the cells in the paracellular pathway, which requires no energy. In general, it increases the drug's molecular size while decreasing the internasal absorption. Thus, the molecular weight of the drug is greater than 1 Da there is a lower blood bioavailability after nasal delivery. These are improved by some absorption enhancers of these substances. Using an absorption enhancer, the recombinant hirudin-2 (rHV2) systemic absorption was highly improved in the nose [11,12].

Barriers interfering nasal drug delivery

Mucus: Mucin, the main amino acids found in mucus, attached to active particles, preventing medication diffusion. Tiny neutral particles can readily move through this barrier, whereas big, charged particles may have difficulty crossing it. Before reaching the respiratory tract's olfactory receptors and mucus, drugs or other foreign substance must penetrate into the water mucus layer of the olfactory region [13,14]. As a result, nasal mucus absorbs the drug is determined by the drug's solubility in mucus. Lipophilic drugs that can partition into the cell membrane's lipid (bilayer) can easily move through biological membranes via the transcellular route. As a result, after nasal delivery, the lipophilic medication rapidly penetrates both the cerebrospinal fluid and the circulation. Polar medicines can only penetrate the mucosal layer and cannot penetrate the lipid bilayer [15].

Nasal mucociliary clearance: The principal defensive system for removing foreign materials, germs, and particles from the nose is mucociliary clearance (MCC). In humans, mucociliary transit has a half-life of roughly 12 to 15 minutes. The most essential components in the Nasal mucociliary Clearance are cilia density, periciliary fluid, and mucus composition. Drug absorption via the nasal epithelium is lowered as a result of this mechanism. By including mucoadhesive substances into the formulation, which may improve nasal absorption, it is possible to overcome the mucociliary clearance and increase the certain period of medications in the nasal region [16].

Enzymic barrier: The lower respiratory airways are protected from dangerous substances by the enzymic barrier of the nasal mucosa. The nasal mucosa barrier, on the other hand, is principally responsible for lowering peptide and protein absorption. Carboxyl esterases, glutathione S-transferases, and cytochrome P450 isoenzymes all of these proteins are found in nasal cells and are involved in drug breakdown within the mucosa. [17] The concentration of enzymes in the olfactory mucosa is higher than in the respiratory mucosa. The nasal cavities of animals, has six times the amount of cytochrome P450 found in the respiratory mucosa (The respiratory mucosal metabolisms are higher than the olfactory mucosa). Recent advances in the discovery and description of several nasal transformation of bio enzymes in animal models, the function of the human nasal mucosa remains unknown. [18]

Factors affecting nasal drugs delivery

Before developing intranasal formulations for brain site, both local delivery, and systemic distribution of drugs, a number of parameters should be considered. The medicine's physicochemical properties, excipients to be used in the development and the nasal cavity physiology state are all listed here. [19,20]

Physicochemical properties of the formulation

pH: Both ionization constant and hydrogen-ion concentration of the formulation, which should be between 4.5 and 6.5, have an impact on the drug's nasal absorption and penetration. When the pH of the formulation was altered to less than 3 and more than 10, intracellular damages were discovered.

Viscosity: Higher the viscosity of the development, time spent in residence of nasal cavity also goes higher and it may also increase the permeation.eg nasal delivery of insulin, metoprolol.

Osmolarity: There will be nasal epithelium shrinkage, when the isotonic solutions are taken in the nasal cavity. Thus, it may cause the increase the drug permeation.

Buffer capacity: The nasal drugs are prepared in smaller quantities, ranging from 25 to 200 liters, and as a result, the pH of the formulation has a significant impact on the concentration of medication (nonionized form) for absorption. As a result, it's critical to keep the formulation's buffer capacity high in order to keep the pH stable.

Mucoadhesive polymer drug delivery platforms

For mucoadhesive bonding to achieve high degrees of retention at applied and targeted areas, polymeric properties such as water liking drugs, negatively charge potential, and hydrogen bond generating groups are required. [21] Furthermore, the polymer's surface free energy must be sufficient to accomplish 'wetting' with the mucus layer. The polymer should also be biocompatible, nontoxic, and cost-effective in order to infiltrate the mucus network. According to Park and Robinson, the polymers commonly used in the creation of mucoadhesive drug platforms that attach to mucin–epithelial interfaces [22].

Reason for choosing polymer for nasal drug delivery: Polymers are important in the advancement of drug delivery technology because they allow for repeated dose, coordinated release of both hydrophilic and hydrophobic medicines, and consistent release of formulations at prolonged time. Biodegradable and bioabsorbable polymers are a fantastic option for a variety of innovative medication delivery methods [23]. A polymeric drug delivery system is a development or technology that allows a therapeutic component to be introduced into the body. Controlling the pace, duration, and location of pharmaceutical release in the body improves the treatment's safety and effectiveness. Medication administration has come a long way in the last two decades, but controlling medication entry into the body, particularly the brain, remains a difficult task. Recent improvements in nano-drug delivery system carrier-mediated transportation across the BBB are starting to provide an origin for targeting medication administration to the brain. Uptake transporters transfer natural nutrients such as amino acids, peptides, hexose, mono-carboxylate, and stem cells across the blood brain barrier [24]. Polymers improve biopharmaceutical pharmacokinetic and pharmacodynamic properties in a variety of ways, including increasing plasma half-life, decreasing immunogenicity, increasing biopharmaceutical stability, low molecular weight drugs which increases the solubility and allowing for targeted drug administration. Rheumatoid arthritis, diabetes mellitus, hepatitis B and C viruses, cancer cells, and ischemia have all been treated with polymer conjugates [25].

Various polymers in nasal drug delivery

Polymers like cellulose derivatives, polyacrylates, starch, gelatin, phospholipids, chitosan, have been found to increase intranasal absorption of the aforementioned therapeutic substances. The recommendation restricts the use of inactive additives with certain of these polymers. These polymers work by either extending medication residence time in the nasal canal or enhancing therapeutic agent intranasal absorption; some polymers may do both. The majority of these polymers are GRAS pharmaceutical excipients [26].

Chitosan

Chitosan is composed of β (1→4)-linked 2-acetamido-2-deoxy-β-D-glucose (N-acetylglucosamine). Chitin is structurally identical to cellulose, but it has acetamide groups (-NHCOCH3) at the C2-portion. On the other hand, chitosan is a linear polymer formed by α (1→4)-linked 2-amino-2-deoxy-β-D-glucopyranose and derived by N-deacetylation, characterized by the degree of deacetylation, which is the copolymer of N-acetylglucosamine and glucosamine. It's a mucoadhesive polymer that's both biocompatible and biodegradable. Chitin is the world's second most prevalent polysaccharide after cellulose. It is hydrophilic due to the presence of a basic amino group, but its solubility and shape in an aqueous media are determined by the number of acetylated monomers and their distribution [27-29]. Chitosan has high mucoadhesive qualities, allowing for paracellular permeability via improving the opening of intestinal epithelial tight junctions. Chitosan and sialic acid's main amino functional groups interact ionically with mucus sulfonic acid resulting in mucoadhesion. [31-33] Furthermore, hydroxyl and amino groups of chitosan and mucus forms the hydrogen bonds in the center of them. The chitosan molecules contain information regarding flexibility, which can be used to increase mucoadhesive properties. This is useful for regulating the drug's release rate [34,35]. Chitosan has a variety of commercial and biological applications. It is primarily used in agriculture as a seed treatment and biopesticide, as plants can easily overcome fungal infections when using it. It is utilized in the pharmaceutical sector for self-healing polyurethane paint coating and also aids in the delivery of medications via the skin [36,37].

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Mucoadhesive polymers classification

Generally, it is classified into mainly two types, which are listed below;

1. Hydrophilic polymers- PVP and methyl cellulose, etc.
2. Hydrogels
✓ Basis of origin
– Natural polymers- chitosan, gelatin and sodium alginate
– Synthetic polymers- cellulose derivatives and carbopol, etc.
✓ Basis on the charge
– Anionic polymers- Carbopol and sodium alginate..
– Cationic polymers- chitosan
– Neutral polymers- eudragit analogues.

Applications of polymers in nasal delivery

As a result of environmental changes, the number of people suffering from nasal allergies has increased, prompting the development of prophylactic and therapeutic pharmaceutical formulations. [38] Because nasal spray dose forms are simple to administer and provide quick relief from nasal symptoms, they have been frequently used for allergic disorders. Drugs have been deposited in the nasal cavity using gel-providing chemicals such as Carbopol-PEG, methylcellulose, and HPMC. [39,40] Any viscous polymer that is swiftly released and then fast vanishes from the nasal canal makes it difficult to postpone medication release, and viscous preparations are very hard to deliver in the nasal cavity [41].

CONCLUSION

In recent years, both biodegradable and biocompatible polymers have been studied for their drug delivery uses in preclinical and clinical studies, resulting in a large increase in polymer-based formulations. The drug administration through nasal route is becoming more common for both local and systemic delivery of therapeutic agents since it is a very expensive and patient-friendly alternative to inject able pharmaceuticals development. Nasal delivery is effective for delivering polypeptides, proamino acids, vaccines, and curing of CNS disorders, as well as peptides, proproteins, and vaccines. A thorough grasp of these challenges is needed for proper polymer that should be prefer in nasal development. The nasal cavity can be used to deliver drugs to treat a range of ailments. Some of the technologies are used to design drug carriers that improve both delivery and performance by using appropriate nasal instruments and increasing drug permeation through the mucosa of the nose. The correlation between the formulation development and the drug delivery devices gives a better drug delivery system to the nose. Boosting the efficiency of this route for the distribution of a variety of drug molecules for the treatment of a variety of ailments. Alternative methods of medication administration are becoming a more prominent topic of study nowadays. Because of their large molecular weight or sensitivity, many recently produced medicines have limited absorption when administered orally. Nasal medication delivery may be a viable strategy. The nose is good accessible, which has adequate blood flow, and, most importantly, provides an easy way to passes the BBB. The process is easy and painless.

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RESEARCH ARTICLE

Genetic and Epigenetic Variations of SARS-CoV-2 and the Hosts: Implications in Humans and Animals

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ABSTRACT

The global COVID-19 pandemic claiming global spread continues to evolve. New episodes of outbreak are possibly caused by the novel variants of concern of severe acute respiratory syndrome corona virus-2 (SARS-CoV-2). The test positivity rate (TPR) and case fatality rate (CFR) have increased steeply in the second wave of COVID-19 compared to the first. From the example of Kerala, a state in southern India, positivity increased from 1.33% at the peak of wave one in 10th June 2020 to 13.45% during 10th June 2021 in the second wave of pandemic. SARS-CoV-2 is an enveloped single-stranded RNA virus. Angiotensin-converting enzyme-2 (ACE-2) is a trans membrane surface protein present on multiple types of cells in the human body to which the viral spike protein attaches. Genetic variations in the SARS-CoV-2 and ACE2 receptor can affect the transmission, clinical manifestations, mortality and the efficacy of drugs and vaccines for COVID-19. Given the high TPR and CFR, it is necessary to understand the variations of SARS-CoV-2 and cellular receptors of SARS-CoV-2 at the molecular level. In this review, we summarize the impact of genetic and epigenetic variations in determining COVID-19 pathogenesis and disease outcome.

Keywords: SARS-CoV-2; COVID-19; TPR; CFR; Genetics; Epigenetics; Sequence variation

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INTRODUCTION

Until recently, human corona viruses have been considered to be insignificant pathogens, which causes mild respiratory illness [1-2]. The first lethal coronavirus outbreak in 2003 (severe acute respiratory syndrome-SARS) alerted global medical community of the challenges faced by coronaviruses. The causative agent SARS-CoV (SARS-corona virus) showed zoonotic origins (from bats) and caused case fatality rate of 10% [3]. High mortality was seen in aged patients and those having co-morbidities [4]. Air-borne infection was also documented in SARS [5,6]. After a decade, in 2013, another coronavirus from dromedary camels, MERS (Middle East respiratory syndrome (MERS) spread to humans in Arabian Peninsula [7]. The virus was named MERS corona virus (MERS-CoV) and the CFR was 20% [8]. The MERS-CoV had ability to alter immune response [18]. In 2017, World Health Organisation (WHO) included SARS and MERS in the priority pathogen list. In December 2019, COVID-19 was initially identified as a novel corona virus (2019-nCoV)–infected pneumonia (NCIP) that was reported from Wuhan, Hubei Province, in China [9]. At the initial stage, the disease was characterised by an incubation period of 5.2 days and a reproductive number estimate of 2.2 [1]. The genomic sequence of the virus was available in early 2020 from the samples isolated from patients suffering from pneumonia in Wuhan, China [10,11]. The SARS-CoV-2 had more amino acid homology with SARS-CoV than MERS-CoV [12].

The WHO announced in January 30, 2020 that the novel corona virus pneumonia epidemic caused by SARS-CoV-2 was classified as a public health emergency of international concern [13]. The international committee on taxonomy of viruses (ICTV) renamed 2019-nCoV as severe acute respiratory syndrome corona virus 2 (SARS-CoV-2) in February 11, 2020 [14]. Since the outbreak of SARS and MERS, COVID-19 is counted the seventh corona virus disease in humans [15]. Although SARS-CoV-2 has lower CFR compared to SARS and MERS viruses, the highly contagious nature of SARS-CoV-2, with an estimated reproduction number (Ro) of 2-6.47 makes COVID-19 a public health concern [16]. The successive emergence of SARS-CoV-2 variants with highly variable pathogenicity and transmission potential warrants studying the genetic diversity and evolution of SARS-CoV-2 with immediate priority [17]. COVID-19 disease dynamics, measured in terms of CFR, TPR and R0, are evolving continuously over time in different geographical locations [18,19]. This differential disease dynamics over time and space could closely be linked to rapidly evolving genetic variations in SARS-CoV-2, interacting with diverse genetic variability of hosts in different areas [20]. Ribosomal frameshifting in corona viruses enable the virus to adapt to the host cell [21].

Bats are considered the reservoir of corona viruses [22]. Wild and domestic animals act as intermediate hosts and facilitate mutation and recombination which enhances genetic diversity of viruses [23]. The coronavirus comprises of two open reading frames (ORF) ORF1a and ORF1b), four structural proteins, and several accessory proteins [24]. Based on the ORF1a and b, coronaviruses are divided into four groups: two mammal-specific alpha and beta and two avian-specific gamma and delta [25]. Inside the host cell, SARS-CoV-2 exerts its action by forming short- and long-range RNA-RNA interactomes which facilitate viral and host RNA interactions [26]. Based on RNA interactome analysis of SARS-CoV-2, 17 host and viral proteins that exert antiviral activity and 9 proviral host factors hijacked by SARS-CoV-2 were identified [26]. This RNA-RNA interaction through RNA binding proteins (RBPs) enables SARS-CoV-2 to evade host immune barrier. Seven human coronaviruses- SARS-CoV, MERS-CoV, SARS-CoV-2, HCoV-229E, HCoV-OC43, HCoV-NL63, and HCoV-HKU- mainly differ in their accessory proteins. SARS-CoV-2 is one of the RNA viruses with the largest genome. The reference genome of SARS-CoV-2 is 29903 bp single-strand RNA (accession number NC045512, severe acute respiratory syndrome coronavirus 2; isolate Wuhan-Hu-1). There are 630,559 SRA (sequence read archive) runs and 570,941nucleotide records of the SARS-CoV-2 genome in national center for biotechnology information (NCBI, accessed on 07-06-2021). The variants are designated by comparing the genetic sequence of emerging SAR-CoV-2 with that of the reference genome [27,28].

Spike protein in SARS-CoV-2
Spike proteins help the virus to bind with host cells and thus determine the virus-host interactions [29]. For SARS-CoV-2, the most prominent region susceptible to the mutation is the spike (S) protein [30]. Spike protein consists of

two subunits-S (receptor binding subunit) and S1 (membrane fusion subunit). Among these subunits, the S subunit is highly variable, while S1 is conserved compared to S. The S subunit functions as attachment sites that enable the virus to bind to the ACE-2 receptor located on the surface of the host cell [31]. Once the virus binds to the host cell, the S1 subunit helps the fusion of the virus with the host cell membrane. Spike protein has an N-terminal domain (NTD), a C-terminal domain (CTD), and a receptor-binding domain (RBD). The RBD contains receptor binding motifs (437-508) which functions as host receptor-binding residues. The sites involved in host-cell binding and/or host immunity invasion are potential mutation hotspots in SARS-CoV-2 [29].

Host cellular receptors

Immunological competency is the key in determining COVID-19 transmissibility and pathogenicity as infection-preventing immunity disappears earlier than pathogenicity alleviating immunity with a projected outcome of a potential variant of SARS-CoV-2 that may cause acute disease in children [32]. Based on this reasoning, these scientists suggest loss of virulence of SARS-CoV-2 only to cause common cold except in children when COVID-19 spread to an endemic phase. The host cellular receptors differ among corona viruses [33]. For SARS-CoV 1 and SARS-CoV 2, RBD binds to ACE-2 receptors on human host cells. For other human coronaviruses like HCoV- HKU-1 and OC 43, the NTD recognizes sugar derivatives on cell surface while HCoV-229E uses RBD to bind with aminopeptidase N (APN) on human host cells. However, the mouse hepatitis coronavirus uses NTD to bind with carcinoembryonic antigen-related cell adhesion molecule 1 (CEACAM1- the host protein present on the cell surface). In MERS (middle east respiratory syndrome) the viral RBD binds to dipeptidyl peptidase 4 (DPP4) on the host cell surface [29].

SARS-CoV-2 variants

Viewing form this light, common locally evolved variants of SARS-CoV-2 are considered to be originated by the repetitive changes of the structural motifs which are specific to corona virus lineages [34]. These variants are also demonstrated in animal models [35]. In the Italian Sardinia Island, four different clusters of viruses were observed, which were characterised by amino acid substitutions [36]. The probability of mutations in S protein, particularly in NTD and RBD, vary widely as these residues are more flexible in their structural conformation, are functionally unique, and are important in the virus-host cell-binding process. Among NTD and RBD, the latter is more prone to mutations. Although SARS-CoV-2 RBD bind to the ACE-2 cell surface receptors on human cells, the binding target of NTD is yet unknown. The mutations in S protein affect the infectivity of SARS-CoV-2. For example, D614G mutation in the spike protein of SARS-CoV-2 increased the viral infectivity [37]. The SARS-CoV-2 variant identified in southeast England in September 2020, known as B1.1.7 or 501Y.V1 contains eight mutations in the spike gene in addition to the D614G mutation- including two deletions in the NTD (ΔY144 and ΔH69/ΔV70), one substitution in the RBD (N501Y) and another substitution close to the furin cleavage site (P681H). The second variant identified in the Eastern Cape, South Africa is known as B.1.351 or 501Y.V2. Along with the D614G mutation, this variant has nine mutations in the spike gene including those in the NTD (R246I and Δ242–Δ244), RBD ((N501Y E484K and K417N), and one mutation close to the furin cleavage site (A701V). The third variant known as P.1 or 501Y.V3 was identified from Brazil and contains three substitutions at RBD (N501Y, E484K, K417T). These mutations in the new variants of SARS-CoV-2 could affect the treatment with monoclonal antibodies and may affect the protective value of vaccines [38].

ACE-2 receptor protein

Genetic variability of host receptor proteins also influences the susceptibility, pathogenicity, and disease progression of viral diseases [39]. As ACE-2 receptor protein is the entry point and site of attachment of SARS-CoV-2, the genetic variability of ACE-2 is considered as a risk factor in the COVID-19 pandemic. The ACE-2 gene is located on the Xp22 chromosome and possesses 22 exons. The expression level of ACE-2 varies with individuals and within an individual, ACE-2 expression is organ-specific. Human, bat (Chinese horseshoe), pig, and civet ACE-2 bind with SARS-CoV spike protein, whereas mouse ACE-2 do not bind with SARS-CoV. Three S protein trimers bind with two ACE-2 dimers initiating the entry of the virus into the host cell. Three amino acid residues in ACE-2 are specifically

involved in SAR-CoV-2 binding. ACE-2 mutations that affect the circulating levels of ACE-2 in individuals are identified. These alleles may predispose varied binding of SARS-CoV-2 to the host cells and variations in COVID-19 pathogenicity. For example, ACE-2 variant rs2106809 was found to be associated with circulating ACE-2 levels. ACE-2 expression also varies with age, sex and ethnicity. Invariably, COVID-19 severity varies among different ethnic groups such as Asians, Africans, and Caucasians. Most of these deviations are explained based on the variations in the allele frequency of eQTLs in diverse populations. Molecules that block angiotensin receptor are suggested against COVID-19 [40].

Other host proteins

Pattern recognition receptors (PRRs) are proteins that control the transcription of inflammatory genes and regulate intracellular signaling processes, and thus, are involved in virus-induced inflammatory responses. Intracellular DNA sensors, RIG-like receptors (RLRs), C-type lectin receptors, (CLRs), Toll-like receptors (TLRs), and NOD-like receptors (NLRs) are major PRRs found in humans. PPRs play important role in innate immune response as they identify PAMPs (pathogen-associated molecular patterns) and DAMPs (damage-associated molecular patterns- molecules released from damaged cells). Overexpression of PRRs, particularly TLR 4 is associated with cytokine storm-mediated morbidity and mortality. The age-related variation in COVID-19 severity is also attributed to differential expression of PRRs. Thus, PRRs play a critical role in mediating SARS-CoV-2-host cell interactions, particularly in individuals with comorbidities like diabetes, obesity, and cardiac diseases [41-42].

Significance of studies on genetic and epigenetic variations

In the rapidly spreading phase of COVID-19, it is very important to identify the sequence variability if any, in the circulating viruses and identify the emergence of new variants [43]. It is also important to corelate the implications of current preventive and therapeutic measures to genetics and genomics of COVID-19 [44]. For example, Pachetti *et al*. (2020) evaluated impact of lockdown on viral mutations and COVID-19 CFR in various countries in Europe and North America. They found that mutations in SARV-COV-2 tend to stabilise at four genomic regions of SARS-CoV-2 whereas, new nonsynonymous mutation was seen in the samples from Sweden where soft lockdown was implemented [45]. In fact, virus surveillance through genomic sequencing helps to understand the mutations in the virus genome and helps to formulate control/therapeutic strategies. Escape variants (the mutated virus that causes disease in vaccinated or recovered individuals) should also be monitored promptly. Exploring genetic variations in the host cell receptors may also help us to elucidate the mechanistic ways by which SARS-CoV-2 differentially bind to the host cells and cause differential susceptibility and pathogenicity in different individuals/diverse populations [46].

Epigenetic changes

There are reports showing disproportionate clinical manifestation of COVID-19 in old age people. Involvement of differential abundance and activity of 315 miRNAs common for age related signaling and COVID-19 are identified and their lower activity is reported in old age and comorbidity conditions [47]. Circular non-coding RNA (CircRNAs- formed by covalent attachment of 3′ and 5′ ends of RNA) and long non-coding RNAs (LncRNAs-with lengths greater than 200 nucleotides) that regulate cell division, cell death, immune response and signaling pathways are involved in COVID-19 pathogenesis. Wu et al (2020) identified 114 circRNAs and 10 lncRNAs in COVID-19 patients that were connected to exosomes. Exosomes are 40-100 nm size vesicles present in the cell which mediate virus-hot cell interaction and influence cellular response against viral diseases. These circRNA and lncRNA are projected as biomarkers to COVID-19 severity [48].

Impact on clinical manifestations

The significance of deletions in immune evasion on the evolutionary trajectory of SARS-CoV-2 to an endemic virus was studied [49]. Deletions in ORF7, ORF8, and ORF10 regions found in Bangladesh and deletions in or near the furin polybasic site of the spike protein have been associated with reduced virulence [50]. The three HLA alleles (HLA-A*11, -C*01, and -DQB1*04) among Spaniards and the HLA-DRB1*08 in the Italian population correlated with

mortality of COVID-19 [51]. Variants in cytokine genes such as IL1B, IL1R1, IL1RN, IL6, IL17A, FCGR2A, and TNF could be related to disease susceptibility, cytokine storm, and other COVID-19 complications [50]. Several variants in ACE2 and TMPRSS2 affecting the expression of the receptors related to COVID-19 have been associated with the disease susceptibility and risk factors. Germline variants in UNC13D and AP3B1 (two typical hemophagocytic lymphohistiocytosis related genes) were found to be associated with the development of severe cytokine storm and fatal outcomes in COVID-19 [52]. Deaths during the first phase of the epidemic was found to be associated with L84S mutation (ORF8 protein involved in immune system evasion) and 2 other helicase mutations (NSP13, P504L, and Y541C) [50-54].

Impact on COVID-19 tests

Rapid and accurate COVID-19 tests are crucial for diagnosis and treatment, prevention, contact tracing, epidemiologic characterization, and public health decision making. The major concern regarding COVID-19 diagnostic tests related to the emergence of new variants is the potential failure of RT-PCR tests for diagnostics. New variants can affect the performance of certain tests if the mutation is in a region of the genome targeted by the test [57]. A high-frequency mutation on the right end of a primer or probe position of a target would possibly produce more false-negatives in diagnostics. The impact of new variants on test sensitivity is influenced by the genomic sequence of the variant, the test design and the prevalence of the particular variant in the tested population. Tests based on multiple regions of the genome may be less impacted by the new variants than the tests that rely on detection of only a single region [58].

Impact on COVID-19 treatment

Several new compounds are tested and some existing drugs are studied for repurposing to target various SARS-CoV-2 proteins [59,60]. For instance, a compound ABBV-744 was found to bind with the main protease enzyme of SARS-CoV-2 with heavy binding affinity (ΔGbin −45.43 kcal/mol) [61]. Viral proteases are also considered drug target against COVID-19 [62]. For advanced lung injury in COVID-19 stem cell therapy using extracellular vesicles is suggested [63]. Variations of TMPRSS2 and CD26 have been demonstrated to enhance COVID-19 susceptibility [64]. Analysing molecular pathways involved in humans also helps to formulate novel treatment protocols. For example, ATP1A1belonging to multi subunit Na+/K+ ATPase is an integral membrane protein responsible for maintaining electrochemical gradients across plasma membrane and transmits cardiotonic steroid binding induced signals in to the cells. ATP1A1 is also involved in the entry of viruses in to the cells, blood coagulation and oxidative stress during COVID-19 [65-66]. Burkard *et al.* (2015) reported that ATP1A1 regulated Srcsignalling blocks entry of corona virus in to host cells [67]. Cardiac glycoside bufalin, that inhibits ATP1A1 is suggested as inhibitor of SARS-CoV-2 [68].

Non-epigenetic drugs re-purposed for treating COVID-19 aiming at the most significant pathways in the SARS-CoV-pathogenesis were not completely successful in eliminating the virus [69]. Recently approved monoclonal antibodies (bamlanivimab plus etesevimab and casirivimab plus imdevimab) for the treatment of non-hospitalized patients with mild to moderate COVID-19, targeting the S protein could limit progression to severe disease particularly in those who have not yet developed an endogenous antibody response [70]. Some SARS-CoV-2 variants (The P.1 variant, B.1.429/B.1.427 variants & B.1.526 variant) have markedly reduced susceptibility to even the newer monoclonal antibodies like bamlanivimab and may have lower sensitivity to etesevimab and casirivimab [71]. Ongoing population-based genomic surveillance of the circulating SARS-CoV-2 variants and their susceptibility to available drugs including anti-SARS-CoV-2 monoclonal antibodies, will be important in defining the utility of the strategy in the future. The molecular level understanding on epigenetic mechanisms including microRNA, DNA methylation, chromosomal structural alterations, and microbiome involvement will provide useful information on the importance of epigenetics in SARS-CoV-2 disease [72]. Epigenetic enzymes (such as DNMT1, histone acetyltransferase 1 (HAT1), histone deacetylase 2 (HDAC2), and lysine demethylase 5B (KDM5B) responsible for the modifications of ACE2 expression including DNA methylation and histone modifications can act as potential targets to control the host immune response [73].

Impact on vaccine efficacy

Neutralising antibodies against SARS-CoV-2 has been evaluated [72]. The experimental data on neutralization assays shows that the plasma from vaccinated individuals has mild to modest reduction in neutralizing activity against mutants. Evidence from clinical trials is broadly consistent with the laboratory results, with the B.1.351 variant showing greater signs of vaccine escape. The ChAdOx1 nCoV-19 vaccine showed clinical efficacy against the B.1.1.7 variant but failed to provide protection against mild to moderate disease caused by the B.1.351 variant, with vaccine efficacy against the variant estimated at 10.4%. The NVX-CoV2373 (Novavax) protein-based vaccine have 95.6% efficacy against the wild-type virus and that this is moderately lower for the B.1.1.7 variant (85.6%) and is further reduced for the B.1.351 variant (60.0%) as per the preliminary data from clinical trials. The JNJ-78436735 (Johnson & Johnson/Janssen) vaccine showed 72% protection against moderate to severe SARS-CoV-2 infections in the USA, but the proportion significantly decreased to 57% in South Africa (at a time when the B.1.351 variant was widespread). Sequencing of viruses associated with prolonged infections will provide useful information on mutations that could contribute to increased escape from vaccine-mediated immunity [74].

CONCLUSIONS AND WAY FORWARD

Genetic characterization of the propagating and /or the emerging variants of the SARS-CoV-2 and identifying the genetic diversity of host cell receptor proteins among distinct populations (e.g., individuals with mild COVID-19 disease vs individuals with severe disease) could offer possible solutions for the management and prevention COVID-19 both in individuals and among different populations. This can be achieved through setting up of centrally co-ordinated local laboratory facilities with genomic variant screening at district levels and linking those with the line departments at the field level.

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RESEARCH ARTICLE

Effect of Reactive Agility Training on Body Control and Cognitive Flexibility in Adolescent Football Players

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ABSTRACT

Football is a team sport and requires rapid Change of direction movements in team sports are not pre planed but performed as a reaction to specific stimulus. Thus requires both body control and cognitive flexibility to give a quick and efficient response with less reaction time on field that is vital for the sports performance. Agility is one such feature of successful athlete. Reactive agility training and change of direction training are training methods to improve the body control and reaction time. To identify the effect of reactive agility training and change of direction drills for 8 week interval in improvement of body control and cognitive flexibility by reducing the time taken to complete the modified Illinois agility test and stroop test in adolescent football players. 40 Participants of age group ranging from 10 to 19 year of age after being screened for eligibility criteria divided into 2 groups by simple random sampling. Group 1 received intervention in the form of reactive agility training along with their regular training and Group 2 received regular training as change of direction training thrice a week for 8 week. Effectiveness Parameters were improvement in modified Illinois agility test and stroop test time. They were assessed at baseline and at the end of the study. Post intervention data was analysed using SPSS software using t test and ANCOVA. There was significant difference in parameters in both the groups from baseline and end of the study($p<0.001$).However reactive agility group was superior to the change of direction control groupThe result of this study demonstrates that reactive agility training and change of direction drills statistically improves the body control in terms of modified Illinois agility test and stroop test. Where reactive agility test showed more improvement than the change of direction drills.

Keywords: Football, Cognitive Flexibility, modified Illinois test, stroop test, reactive agility training.

INTRODUCTION

Successful performances in field sports games like as football, basketball, and soccer demands crisp recognition and quick reaction to situations on field. Reaction to the opponent as well as the team mate is required to perform well on field. Sports performance in football can be assessed by observing the physical conditioning of the player, his/her technical skill and tactical skills during the game. Thus proves the need of specific sports training in the athletes for better performances. Sports Training involves a systematic development of each component of sports performance and is dependent on the time period of the training. So as the players age their seniority level increases and thus they achieve maximum efficiency in the performance through experience level. Thus emphasizing specific sports training [2]. The specific components of sports training are 1) Physical that includes the somatic and fitness component.2) Technical component including Fundamental skills and Sports skills. 3) Tactical components whose key features are Strategy and Tactics.In which strategy is pre planned where as tactic is a practical execution of the strategy in response to a specific game situation.4) Psychological component.

Match of football require maximal acceleration and fatigue resistance in terms of endurance. The sprints can be in single direction or multi direction and requires frequent abrupt turning, twisting and explosive change of direction movements in response to the opponent player that is reactive agility. Parameters like Acceleration and velocity are highly related to the agility. It depends on the biochemical and anatomical structure a muscle constitute especially the type of fibres. According to Sporis (2010), Agility is the ability to maintain and control the position of the body when changing direction quickly and precisely without losing his balance while on the move. Where Sheppard & Young [9] proposed that agility is: "a rapid whole body movement with a change of velocity or direction in response to a stimulus". This definition considers both the physical and cognitive parameters that contribute to a successful agility performance [11,12]. Thus he developed a reactive agility test (RAT) using a real person (human stimulus) performing some movement patterns for subjects to react to in the same direction as the human stimulus [14]. Cognitive functions, processing of vision and timing of its perception. Whereas Anticipation, which cannot be pre-planned are part of reactive components. Thus young players should be trained for the executive component. Executive function is the other name for cognitive control. The main pillars of the executive functions are 1) Inhibition and interference control 2) Working memory 3) Cognitive flexibility.

Other influencing factors are pattern recognition and memory of movement pattern. The readiness of a player to adapt to stimulus from environment is also known as the cognitive flexibility. The athlete being able to anticipate where the ball is going next is the cognitive skill and such movements are studied under the umbrella of performance improving agility. Study has recognized agility as a trainable motor skill that can be achieved through proper progressive practice [4]. That means Exercises that includes change of direction, stop, and move abruptly, which in turn will increase the agility to rapidly and efficiently decelerate and accelerate in an effort to react appropriately to task-relevant cues during the game and should be trained and speed [5]. Reactive Agility Training (RAT) is an exercise to improve agility and speed. The used of RAT is suggested to improve an athlete ability to read and react to stimuli, which in turn may increase the aspects of agility performance (Holmberg, 2009), by improving anticipatory and/or decision-making responses to movement (Sheppard et al., 2006).

Thus to train choice reactions Reactive agility drills should be constitute in the specific filed training to make the nervous system learn, how to control the body on reacting to a stimulus. It will also help to correct body position, balance, coordination and explosive movement pattern [5]. So this study focus on the effect of reactive agility training on the physical as well as cognitive aspect of the agility. To test the body control after the reactive agility training we will be employing the modified part of the Illinois agility test. Any improvement in the cod drills would indicate transfer of training effect and may facilitate potential improvements in sports performance.

Also some studies suggest that reactive agility is used to differentiate the players on basis of their performance level in soccer players, as the reactive agility is greater in the elite professional and senior players than younger beginner

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players. Thus training adolescents for reactive agility may help them to reach the elite level at young age Thus with combination of the Illinois agility test and stroop test we aim to find out the effect of reactive agility training program of 8 weeks on the parameters like body control and cognitive flexibility respectively.

AIM AND OBJECTIVE

This study aims to identify and compare the effect of reactive agility training and change of direction drills on body control and cognitive flexibility in 8 weeks duration in adolescent football players in terms of scores of modified Illinois agility test and stroop test

MATERIALS AND METHODOLOGY

SOURCE OF DATA: Kahani football club and Kahani sports Academy, Ahmedabad
STUDY DESIGN: experimental study
SAMPLE SIZE: 40
SAMPLE METHOD: Simple Random Sampling
DURATION: 1 YEAR
TRAINING DURATION: Alternate days thrice a week for 8 weeks.

The study was conducted over a period of 8 weeks. A total of 40 participant of age group ranging from 10 to 19 years of age were screened for eligibility criteria and divided into two groups by simple random sampling. Group 1(n=20) received reactive agility training along with their regular training and group 2(n=20) received their regular training in the form of change of direction drills. Elite player or players with Leg injuries like sprain, strain, fracture etc in last 6 months. Players with neurological or cognitive dysfunction, Hearing difficulties or use of aids or Colour blind individual were excluded. Effectiveness parameters were improvement in modified Illinois agility test and stroop test time. They were assessed at baseline and at the end of the study.

PROCEDURE

All subjects were asked to keep their activity level stable. During the training period they were asked to log in the activity log if any additional exercise or activity apart from the training were undertaken. This study used the Encephal App-Stroop (IC=0.83) [11]. It is a Smartphone application based on the adaptation of the classical test. It has a colour and word test in which the subject reads the name of randomly placed colours and must always select the colour of the sign that appears on the screen .It could be (green, blue, red , yellow). It has 2 conditions of test:(1) with no interference that is stroop effect off.(2) with interference that is stroop effect on. The other evaluation tool was the modified Illinois agility test. This modified version of Illinois agility test was proposed by Vescovi & Mc Guigan (2008), has the same layout but takes less time as it has less straight sprinting (2 x 10m instead of 4 x 10m) with CI=95% for under 14 soccer players [13].

TRAINING PROTOCOL FOR EXPERIMENTAL GROUP

The training sessions were in phases, they were divided into 4 phases each of two week. Subjects underwent Functional warm up which includes: Walking lunges, Side shuffle, Dynamic stretching, Carioca before the training session

PHASE 1 : it was more of preconditioning with not less than 135 degree cuts in the drills .

BOX CONE DRILLS
Also known as quickness box. A number is called out to the player. He then runs, back paddles and shuffle as needed to touch the cone. To touch the cone he may use either of the hands or the hand which is decided before the drill. After touching the cone he waits while shuffling for the next command.

LADDER DRILLS
It is usually used improve foot work and can also impact performance from changing direction to blocking and tackling .1)One step in each hole 2)Two steps in each hole 3)Bunny hop 4)One leg bunny hop (alternate leg on command)5)Hop scotch 6)From the side 7)Back foot 8)From the side.

PHASE 2: (3-4week): drills with 90 degree cut
CATCHING A REACTION BALL
A gate of cones will be kept between the coach and the athlete. The coach whistles and throws the tennis ball from either right or left hand (not pre planned) and the athlete has to react and sprint toward s the coach on either left or right and catch the ball before the ball falls on the ground.

MIRROR DRILLS
Purpose of this drill is to build the offensive lines mans to side shuttle. Co ordination of speed and reaction time is required for this drill. Defender is asked to run around offensive line man. The offensive line man will be continuously changing direction .change of direction will be in form of moving laterally from side to side throughout the length of drill (10 m) .The defender must mirror movement of the offensive line man and stay in front of him with lowered body and shuffling feet.

PHASE 3 (4-6 WEEK)
In this phase more advance training will be done. Progression of the ladder drills and more emphasis on acceleration and deceleration of the player.
1) QUICK, RISE AND GO:the athlete stands at the starting point of the sprint setup. Once the coach calls QUICK the athlete shuffles at place, as the coach calls raise the athlete raise his hands, and when the coach calls go he accelerates from his position. This way it works on the reaction time.
2) FORWARD TO BACKWARD SHUTTLLE:The athlete shuffles forward and backward between cones for 3 sets of 3 reps
3) CIRCLE SHUFFLES TO ACCELERATION:The commands will be given by the coach from which side the athlete has to shuffle either right or left.
4) Single leg plant to 20 yards sprint:The athlete side jumps and plants a single limb and the sprint forward for 20 yards.

PHASE 4(7-8 WEEK)
more explosive training by modifying with more complex drills by making game like situation by adding ball in the drills, reacting to partners ,quick change in commands, using interference patterns by showing colour cards to signal to run to left and right and changing them in between to make the task more complex etc.

STATISTICAL ANALYSIS
All pre and post data were primarily analyzed with Shapiro Wilk test where most of the parameters showed the normal distribution. So Parametric tests were used. Paired and unpaired t test was employed to analyse the difference between groups and within the groups with level significance set at p<0.001.

RESULTS

Table 1 and 2 shows paired t test for both the groups. Both the groups shows statistically significant result pre and post intervention with p<0.001 on paired t- test.

DISCUSSION

Statistical significant difference was found in both the group pre and post test all the three parameter showed significant improvement in the experimental group that received the reactive agility training. Whereas change of direction training also showed difference as an improvement but its mean difference was less than that of the reactive agility group. The improvement in the stroop test both congruent and incongruent that is stroop off and stroop on time suggests that decision making power and reaction speed has an impact of the reactive agility training. Thus we reject the null hypothesis that there is no significant difference in the group pre and post training as the cognitive flexibility has significantly increased as the stroop test time reduced post intervention. The other parameter that is the body control that has also showed a significant improvement in both the training group the change of direction group as well as the reactive agility group. The experimental group that received reactive agility training along with their regular training had shown statistically significant result. Thus it is clear that both the agility training be it the only pre planned change of direction training or the open circuit reactive agility training has a positive effect on the physical as well as the cognitive aspect of fitness.

In addition to the physical benefit of AT, it may also improve cognitive performance animal studies on this subject suggest that both running and agility exercise increase the neurogenesis in hippocampal area of brain [27], thus spatial navigation and memory power is increased. Whereas some study propose that Agility exercise also results in synaptogenesis in the motor cortex and cerebellum .whereas running alone may not. Concluding that AT positively affect a number of regions of the brain and that may be the reason that influence the players cognitive function [27] On assessment of the activity log at the conclusion of the training we found no study protocol training related injuries in both the group during training. Thus as an secondary outcome to the result of the study we conclude that training adolescent beginner players in phasic manner along with proper conditioning we can prevent from injuries and proper knowledge of biomechanical comp[nents of movement and their patterns in early childhood it is plu point for future performances. Although this was not an expressed purpose of our protocol. We hereby mention it to encourage future researches on this topic to consider including an injury comparisons as a priority measure.

Training the adolescent football players at their pubertal or pre pubertal age that is the growing age and age of development i indicated beneficial in accordance to the study of Van der Fels et al [27]. He discussed the association between motor skills and cognitive function in children and indicated inconsistent associations and observed a weak to strong association between some motor skills and underlying cognitive skills tests that suggested the complexity of motor skills has an association. Studies suggest that there is a strong relation of motor and cognition in pre-pubertal than pubertal -children. Circumpubertal and postpubertal adaptations are likely to be mediated by increases in sex androgen concentrations such as testosterone, growth hormone, and insulin-like growth factor [26]. Hormonal changes during puberty increases force-producing abilities. That acts as cherry on the top of the cake for continued neural development. It can also be efficient in increasing muscle cross-sectional area. For that it is vital to know the muscle pennation angle and continued fiber and its type differentiation. It can be observed that children and adolescents gains strength. Thus, for the improvement of change of direction skills technical as well as physical fitness qualities are necessary.

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CONCLUSION

Thus the study concludes that reactive agility training improve s body control and promotes cognitive flexibility in adolescent football players when compared to the conventional change of direction training in 8 week duration.

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Table1 : Shows Comparison Between the Groups by Unpaired T Test

Comparison Between Groups						
OUTCOMES		Mean	Std. Deviation	T	p Value	Sig/Non Sig
Difference stroop on	Experimental	1.77	0.96	4.89500	<0.001	Sig
	Control	0.59	0.42			
Difference Stroop Off	Experimental	1.82	0.41	7.75200	<0.001	Sig
	Control	0.54	0.57			
Diff illinois agility test	Experimental	12.00	3.40	5.40900	<0.001	Sig
	Control	5.32	4.07			

There is a statistically significant difference between the groups with unpaired t-test.

Table 2 Shows Paired t test

Group 1 (experimental)	Mean	SD	t	P Value
Pre Stroop off	3.29	0.73	18.88	<0.001
Post Stroop off	1.47	0.61		
Pre Stroop on	3.00	0.96	7.84	<0.001
Post Stroop on	1.23	0.21		
Pre Illinois agility test	25.67	3.76	14.99	<0.001
Post Illinois agility test	13.67	2.25		

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Group 2 (control group)	Mean	SD	t	P Value
Pre stroop off	3.31	0.66	4.13	<0.001
Post stroop off	2.76	0.58		
Pre stroop on	3.10	0.99	6.12	<0.001
Post stroop on	2.51	0.83		
Pre Illinois agility test	22.32	2.67	5.69	<0.001
Post Illinois agility test	17.00	5.10		

Table 3 : ANCOVA for stroop off variable

Stroop Off						
Adjusted Post Test Means		Sum of Squares	df	Mean Square	F ratio	p value
Experimental	Control					
1.475	2.758	15.214	1	15.214	84.42	<0.001

Shows significant result with p<0.001.

Table 4: ANCOVA for stroop on variable.

Stroop On						
Adjusted Post Test Means		Sum of Squares	df	Mean Square	F ratio	p value
Experimental	Control					
1.253	2.489	14.076	1	14.076	64.493	<0.001

Shows significant result with p<0.001.

Table 5. ANCOVA for Illinois agility test variable.

ANCOVA: Illinois						
Adjusted Post Test Means		Sum of Squares	df	Mean Square	F ratio	p value
Experimental	Control					
12.666	17.948	201.12	1	201.12	15.917	<0.001

Shows significant result with p<0.001.

RESEARCH ARTICLE

Few Inequalities of Fermatean Fuzzy Matrices

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ABSTRACT

Certain inequalities of Fermatean fuzzy matrices in connection to algebraic sum and product are presented in this study.

Keywords : Fermatean fuzzy matrix, Algebraic Sum, Algebraic Product, commutative monoid

INTRODUCTION

By inventing the Fuzzy set in the decision-making process, Zadeh[1] modelled inaccurate evaluations. The intuitionistic fuzzy set (IFS) is a variant of the fuzzy set (FS). The degree of membership and non-membership are its distinguishing features. This was devised by Atanassov[2] to ensure that the sum is either less than or equal to 1. Thomason[3] was the first to introduce fuzzy matrices. In that case, he's talking about the convergence of fuzzy matrix powers. It has a significant impact on scientific progress. The fundamentals of fuzzy matrix theory are also covered. The authors primarily attempted to highlight the fundamental requirements for improving the fuzzy model. The authors do not use any fine prepositions to shape the fuzzy matrices. Instead, they provided the most basic characteristics along with example methods. Some writers proposed the convergent power sequence of fuzzy matrices because it produced good results. Yager[4,5,6] later suggested the concept of Pythogorean fuzzy set (PFS). The membership and non-membership degrees both meet the requirement that the sum of the forecourt is less than one. Tapan Senapathi and Ronald R.Yager[7] proposed Fermatean fuzzy sets as an extension of Pythagorean fuzzy sets. Fermataean fuzzy matrices were first introduced by Silambarasan [8]. The qualities of FFMs having a mix of operations with well-known operations are examined and studied using algebraic properties. This study formulates and proves inequalities associated with the algebraic operations of fermatean fuzzy matrices.

Preliminaries

In this part, we'll go over some basic definitions for fermatean fuzzy matrices.

Definition 2.1:

A Fermatean Fuzzy Matrix (FFM) is a matrix $R = (< \alpha_{r_{ij}}, \beta_{r_{ij}} >)$ which is a set of non-negative real numbers that satisfies $(\alpha_{r_{ij}})^3 + (\beta_{r_{ij}})^3 \leq 1$ for all i, j.

Definition 2.2:

Let $R = (< \alpha_{r_{ij}}, \beta_{r_{ij}} >)$, $S = (< \alpha_{s_{ij}}, \beta_{s_{ij}} >)$ be two FFMS of same order then

$$R \vee S = \left(\langle \max \left(\alpha_{r_{ij}}, \alpha_{s_{ij}} \right), \min \left(\beta_{r_{ij}}, \beta_{s_{ij}} \right) \rangle \right)$$
$$R \wedge S = \left(\langle \min \left(\alpha_{r_{ij}}, \alpha_{s_{ij}} \right), \max \left(\beta_{r_{ij}}, \beta_{s_{ij}} \right) \rangle \right)$$
$$R^c = \left(\langle \beta_{r_{ij}}, \alpha_{r_{ij}} \rangle \right)$$

$R \leq S$ if $\alpha_{r_{ij}} \leq \alpha_{s_{ij}}$ and $\beta_{r_{ij}} \geq, \beta_{s_{ij}}$ for all i, j.

The algebraic sum of R and S is defined as

$$R \oplus_F S = (< \sqrt[3]{\alpha_{r_{ij}}^3 + \alpha_{s_{ij}}^3 - \alpha_{r_{ij}}^3 \alpha_{s_{ij}}^3}, \beta_{r_{ij}} \beta_{s_{ij}} >)$$

The algebraic product of R and S is defined as

$$R \odot_F S = (< \alpha_{r_{ij}} \alpha_{s_{ij}}, \sqrt[3]{\beta_{r_{ij}}^3 + \beta_{s_{ij}}^3 - \beta_{r_{ij}}^3 \beta_{s_{ij}}^3} >)$$

Definition 2.3:

If all the entries of FFM are <0, 1> then it is known as zero FFM denoted as 0

If all the entries of FFM are <1, 0> then it is known as universal FFM denoted as J.

Section 3

On Fermatean Fuzzy Matrices, we prove a number of inequalities relating to algebraic operations.

Let $\mathcal{F}_{mn}$ denotes the set of all Fermatean Fuzzy Matrices of order mxn.

Theorem 3.1:

$T \geq R \vee S$ if $T \geq R$ and $T \geq S$ where $R, S, T \in \mathcal{F}_{mn}$

Proof:

If $T \leq R$ then , $\alpha_{r_{ij}} \leq \alpha_{t_{ij}}$ and $\beta_{r_{ij}} \geq \beta_{t_{ij}}$ for all i,j

If $T \geq S$ then , $\alpha_{s_{ij}} \leq \alpha_{t_{ij}}, \beta_{s_{ij}} \geq \beta_{t_{ij}}$ for all i,j

$\max \left(\alpha_{r_{ij}}, \alpha_{s_{ij}} \right) \leq \alpha_{t_{ij}}$ and $\min \left(\beta_{r_{ij}}, \beta_{s_{ij}} \right) \geq \beta_{t_{ij}}$ for all i, j

which implies $T \geq R \vee S$

Theorem 3.2:

$S \vee T \geq R \vee T$ if $S \geq R$ where $R, S, T \in \mathcal{F}_{mn}$

Proof:

If $R \leq S$ then $\alpha_{r_{ij}} \leq \alpha_{s_{ij}}$ and $\beta_{r_{ij}} \geq \beta_{s_{ij}}$ for all i, j

$\max \left(\alpha_{r_{ij}}, \alpha_{t_{ij}} \right) \leq \max \left(\alpha_{s_{ij}}, \alpha_{t_{ij}} \right)$ and $\min \left(\beta_{r_{ij}}, \beta_{t_{ij}} \right) \geq \min \left(\beta_{s_{ij}}, \beta_{t_{ij}} \right)$ for all i , j

For $x, y, z \in \mathbb{R}$ if $x \leq y$ then

$\max (x, z) \leq \max (y, z)$ and $\min (x, z) \leq \min (y, z)$

$S \vee T \geq R \vee T$.

Theorem 3.3:

$T \geq R \wedge S$ if $R \geq T$ and $S \geq T$ where $R, S, T \in \mathcal{F}_{mn}$.

Proof:

If $R \geq T$ and $S \geq T$ then

$\alpha_{t_{ij}} \leq \alpha_{r_{ij}}$ and $\beta_{t_{ij}} \geq \beta_{r_{ij}}$ for all i , j

$\alpha_{t_{ij}} \leq \alpha_{s_{ij}}$ and $\beta_{t_{ij}} \geq \beta_{s_{ij}}$ for all i , j

For $a, b \in \mathbb{R}$ if $\leq b$, $a \leq c$ then $a \leq \min(b, c)$ also $c \geq \max(a, b)$.

Thus $\alpha_{t_{ij}} \leq \min(\alpha_{r_{ij}}, \alpha_{s_{ij}})$ and $\beta_{t_{ij}} \geq \max\left(\beta_{r_{ij}}, \beta_{s_{ij}}\right)$ for all i, j.

So $\left(\min\left(\alpha_{r_{ij}}, \alpha_{s_{ij}}\right), \max\left(\beta_{r_{ij}}, \beta_{s_{ij}}\right)\right) \geq (\alpha_{t_{ij}}, \beta_{t_{ij}})$

$R \wedge S \geq T$

Theorem 3.4:

Let $R, S, T \in \mathcal{F}_{mn}$ and if $S \geq R$ and $T \geq R$ and $T \wedge S = 0$ then R=0.

Proof:

If $R \leq S$ then $\alpha_{r_{ij}} \leq \alpha_{s_{ij}}$ and $\beta_{r_{ij}} \geq \beta_{s_{ij}}$ for all i , j

$S \wedge T = 0 \Rightarrow \min\left(\alpha_{s_{ij}}, \alpha_{t_{ij}}\right) = 0$

$\max\left(\beta_{s_{ij}}, \beta_{t_{ij}}\right) = 1$ for all i , j.

Since $\alpha_{r_{ij}} \leq \alpha_{s_{ij}}$ and $\alpha_{r_{ij}} \leq \alpha_{t_{ij}}$

We have $\alpha_{r_{ij}} \leq \min\left(\alpha_{s_{ij}}, \alpha_{t_{ij}}\right) = 0$

$\alpha_{r_{ij}} \leq 0 \Rightarrow \alpha_{r_{ij}} = 0$ for all i , j.

Since $\beta_{r_{ij}} \geq \beta_{s_{ij}}$ and $\beta_{r_{ij}} \geq \beta_{t_{ij}}$

We have $\beta_{r_{ij}} \geq \max\left(\beta_{s_{ij}}, \beta_{t_{ij}}\right)$ for all i, j.

$\Rightarrow \beta_{r_{ij}} \geq 1$

$\Rightarrow \beta_{r_{ij}} = 1$

That is R=(<0,1>) for all i , j.Hence R=0.

Theorem 3.5:

Let $R, S, T \in \mathcal{F}_{mn}$ and if $R \geq S$ then $R \wedge T \geq S \wedge T$.

Proof:

If $R \geq S$ then $\alpha_{r_{ij}} \geq \alpha_{s_{ij}}$ and $\beta_{r_{ij}} \leq \beta_{s_{ij}}$ for all i , j.

$\min\left(\alpha_{r_{ij}}, \alpha_{t_{ij}}\right) \geq \min\left(\alpha_{s_{ij}}, \alpha_{t_{ij}}\right)$

$\max\left(\beta_{r_{ij}}, \beta_{t_{ij}}\right) \leq \max\left(\beta_{s_{ij}}, \beta_{t_{ij}}\right)$

So, $\wedge T \geq S \wedge T$.

Theorem 3.6:

Let $R, S, T \in \mathcal{F}_{mn}$

(a) $S \vee T \geq R$ if $(S \wedge R) \vee (T \wedge R) = R$

(b) $S \wedge T \leq R$ if $(R \vee S) \wedge (R \vee T) = R$

Proof:

To prove (a) :

$$R = \left(\langle \max\left(\min\left(\alpha_{r_{ij}}, \alpha_{s_{ij}}\right), \min\left(\alpha_{r_{ij}}, \alpha_{t_{ij}}\right)\right), \min\left(\max\left(\beta_{r_{ij}}, \beta_{s_{ij}}\right), \max\left(\beta_{r_{ij}}, \beta_{t_{ij}}\right)\right)\rangle\right)$$

$$= \left(\langle \min\left(\alpha_{r_{ij}}, \max\left(\alpha_{s_{ij}}, \alpha_{t_{ij}}\right)\right), \max\left(\beta_{r_{ij}}, \min\left(\beta_{s_{ij}}, \beta_{t_{ij}}\right)\right)\rangle\right)$$

$$\Rightarrow \min\left(\alpha_{r_{ij}}, \max\left(\alpha_{s_{ij}}, \alpha_{t_{ij}}\right)\right) = \alpha_{r_{ij}}$$

$$\max\left(\beta_{r_{ij}}, \min\left(\beta_{s_{ij}}, \beta_{t_{ij}}\right)\right) = \beta_{r_{ij}}$$

Thus $\alpha_{r_{ij}} \leq \max\left(\alpha_{s_{ij}}, \alpha_{t_{ij}}\right)$ and $\beta_{r_{ij}} \geq \min(\beta_{s_{ij}}, \beta_{t_{ij}})$ for all i , j.

$$\Rightarrow (\alpha_{r_{ij}}, \beta_{r_{ij}}) \leq \left(\max\left(\alpha_{s_{ij}}, \alpha_{t_{ij}}\right), \min\left(\beta_{s_{ij}}, \beta_{t_{ij}}\right)\right).$$

$S \vee T \geq R$

Similarly, we can prove (b)

$$\left(\langle \alpha_{r_{ij}}, \beta_{r_{ij}} \rangle\right) = \left(\langle \min\left(\max\left(\alpha_{r_{ij}}, \alpha_{s_{ij}}\right), \max\left(\alpha_{r_{ij}}, \alpha_{t_{ij}}\right)\right), \max\left(\min\left(\beta_{r_{ij}}, \beta_{s_{ij}}\right), \min\left(\beta_{r_{ij}}, \beta_{t_{ij}}\right)\right)\rangle\right).$$

$$= \left(\langle \max\left(\alpha_{r_{ij}}, \min\left(\alpha_{r_{ij}}, \alpha_{t_{ij}}\right)\right), \min\left(\beta_{r_{ij}}, \max\left(\beta_{s_{ij}}, \beta_{t_{ij}}\right)\right)\rangle\right)$$

$$\alpha_{r_{ij}} = \max\left(\alpha_{r_{ij}}, \min\left(\alpha_{s_{ij}}, \alpha_{t_{ij}}\right)\right) \text{.for all i , j}$$

$$\beta_{r_{ij}} = \min\left(\beta_{r_{ij}}, \max\left(\beta_{s_{ij}}, \beta_{t_{ij}}\right)\right) \text{ for all i , j.}$$

$$\left(\langle \min\left(\alpha_{s_{ij}}, \alpha_{t_{ij}}\right), \max\left(\beta_{s_{ij}}, \beta_{t_{ij}}\right)\rangle\right) \leq \left(\langle \alpha_{r_{ij}}, \beta_{r_{ij}} \rangle\right)$$

$$S \wedge T \leq R$$

Theorem 3.7:

$R \wedge T = 0$ if $S \geq R$ and $S \wedge T = 0$ where $R, S, T \in \mathcal{F}_{mn}$.

Proof:

If $S \geq R$ then $\alpha_{s_{ij}} \geq \alpha_{r_{ij}}$ and $\beta_{s_{ij}} \leq \beta_{r_{ij}} for$ all i , j

By theorem 3.5, $R \wedge T \leq 0 \Rightarrow R \wedge T = 0$.

Theorem 3.8:

$R \leq S$ if and only if $S^c \leq R^c$ where $R, S \in \mathcal{F}_{mn}$

Proof:

If $R \leq S$ then for all i , j, $\alpha_{r_{ij}} \leq \alpha_{s_{ij}}$ and $\beta_{r_{ij}} \geq \beta_{s_{ij}}$

$\alpha_{r_{ij}} \geq \alpha_{s_{ij}}$ and $\beta_{r_{ij}} \leq \beta_{s_{ij}}$ for all i , j.

which implies $S^c \leq R^c$.

Conversely,   Assume that $S^c \leq R^c$

$\beta_{s_{ij}} \leq \beta_{r_{ij}}, \alpha_{s_{ij}} \geq \alpha_{r_{ij}}$ for all i , j

$\alpha_{r_{ij}} \leq \alpha_{s_{ij}}$ and $\beta_{r_{ij}} \geq \beta_{s_{ij}}$ for all i , j.

$\Rightarrow R \leq S$.

Theorem 3.9:

$S \leq R^c$ if $R \leq S^c$ where $R, S \in \mathcal{F}_{mn}$

Proof:

If $S^c \geq R$ then we have $\alpha_{r_{ij}} \leq \beta_{s_{ij}}$ and $\beta_{r_{ij}} \geq \alpha_{s_{ij}}$ for all i , j.

$\beta_{s_{ij}} \geq \alpha_{r_{ij}}, \alpha_{s_{ij}} \leq \beta_{r_{ij}}$ for all i , j.

$(\alpha_{s_{ij}}, \beta_{s_{ij}}) \leq (\beta_{r_{ij}}, \alpha_{r_{ij}})$

$S \leq R^c$.

Theorem 3.10:

If $R, S \in \mathcal{F}_{mn}$ and if $R^c \leq S$ then $S^c \leq R$.

Proof:

If $R^c \leq S$ then $\beta_{r_{ij}} \leq \alpha_{s_{ij}}$ and $\alpha_{r_{ij}} \geq \beta_{s_{ij}}$ for all i , j

$\beta_{s_{ij}} \leq \alpha_{r_{ij}}$ and $\alpha_{s_{ij}} \geq \beta_{r_{ij}}$

$\Rightarrow (\beta_{s_{ij}}, \alpha_{s_{ij}}) \leq (\alpha_{r_{ij}}, \beta_{r_{ij}})$

$S^c \leq R$.

Theorem 3.11:

Let $S, T \in \mathcal{F}_{mn}$.Then (a) $S \vee (S \oplus_F T) = S \oplus_F T$

(b) $S \wedge (S \oplus_F T) = S$.

Proof:

To Prove(a): $S \vee (S \oplus_F T) = \left(\langle \alpha_{s_{ij}}, \beta_{s_{ij}} \rangle\right) \vee \left(\langle \sqrt[3]{\alpha_{s_{ij}}{}^3 + \alpha_{t_{ij}}{}^3 - \alpha_{s_{ij}}{}^3 \alpha_{t_{ij}}{}^3}, \beta_{s_{ij}} \beta_{t_{ij}} \rangle\right)$

$$= \left(\langle \sqrt[3]{\alpha_{s_{ij}}{}^3 + \alpha_{t_{ij}}{}^3 - \alpha_{s_{ij}}{}^3 \alpha_{t_{ij}}{}^3}, \beta_{s_{ij}} \beta_{t_{ij}} \rangle\right)$$

$$= S \oplus_F T$$

To Prove (b): $S \wedge (S \oplus_F T) = \left(\langle \alpha_{s_{ij}}, \beta_{s_{ij}} \rangle\right) \wedge \left(\langle \sqrt[3]{\alpha_{s_{ij}}{}^3 + \alpha_{t_{ij}}{}^3 - \alpha_{s_{ij}}{}^3 \alpha_{t_{ij}}{}^3}, \beta_{s_{ij}} \beta_{t_{ij}} \rangle\right)$

$$= \left(\langle \min\left(\alpha_{s_{ij}}, \sqrt[3]{\alpha_{s_{ij}}{}^3 + \alpha_{t_{ij}}{}^3 - \alpha_{s_{ij}}{}^3 \alpha_{t_{ij}}{}^3}\right), \max\left(\beta_{s_{ij}}, \beta_{s_{ij}} \beta_{t_{ij}}\right) \rangle\right)$$

$$= \left(\langle \alpha_{s_{ij}}, \beta_{s_{ij}} \rangle\right) = S$$

Theorem 3.12:   If $S, T \in \mathcal{F}_{mn}$ then (a) $S \vee (S \odot_F T) = S$

(b) $S \wedge (S \odot_F T) = S \odot_F T$.

Proof: Consider $S \vee (S \odot_F T) = \left(\langle \alpha_{s_{ij}}, \beta_{s_{ij}} \rangle\right) \vee \left(\langle \alpha_{s_{ij}} \alpha_{t_{ij}}, \sqrt[3]{\beta_{s_{ij}}{}^3 + \beta_{t_{ij}}{}^3 - \beta_{s_{ij}}{}^3 \beta_{t_{ij}}{}^3} \rangle\right)$

$$= \left(\langle \max\left(\alpha_{s_{ij}}, \alpha_{s_{ij}} \alpha_{t_{ij}}\right), \min\left(\beta_{s_{ij}}, \sqrt[3]{\beta_{s_{ij}}{}^3 + \beta_{t_{ij}}{}^3 - \beta_{s_{ij}}{}^3 \beta_{t_{ij}}{}^3}\right) \rangle\right)$$

$$= \left(\langle \alpha_{s_{ij}}, \beta_{s_{ij}} \rangle\right) = S$$

To Prove (b): $S \wedge (S \odot_F T) = \left(\langle \alpha_{s_{ij}}, \beta_{s_{ij}} \rangle\right) \wedge \left(\langle \alpha_{s_{ij}} \alpha_{t_{ij}}, \sqrt[3]{\beta_{s_{ij}}{}^3 + \beta_{t_{ij}}{}^3 - \beta_{s_{ij}}{}^3 \beta_{t_{ij}}{}^3} \rangle\right)$

$$= \left(\langle \min\left(\alpha_{s_{ij}}, \alpha_{s_{ij}} \alpha_{t_{ij}}\right), \max\left(\beta_{s_{ij}}, \sqrt[3]{\beta_{s_{ij}}{}^3 + \beta_{t_{ij}}{}^3 - \beta_{s_{ij}}{}^3 \beta_{t_{ij}}{}^3}\right) \rangle\right)$$

$$= \left(\langle \alpha_{s_{ij}} \alpha_{t_{ij}}, \sqrt[3]{\beta_{s_{ij}}{}^3 + \beta_{t_{ij}}{}^3 - \beta_{s_{ij}}{}^3 \beta_{t_{ij}}{}^3} \rangle\right)$$

$$= S \odot_F T.$$

Theorem 3.13:

Let $R, S \in \mathcal{F}_{mn}$. Then (a) $(R \oplus_F S) = S \oplus_F R$

(b) $(R \odot_F S) = S \odot_F R$

(c) $(R \oplus_F S)^c = R^c \odot_F S^c$

(d) $(R \odot_F S)^c = R^c \oplus_F S^c$

Proof: To Prove (a): $(R \oplus_F S) = \left(\langle \sqrt[3]{\alpha_{r_{ij}}{}^3 + \alpha_{s_{ij}}{}^3 - \alpha_{r_{ij}}{}^3 \alpha_{s_{ij}}{}^3}, \beta_{r_{ij}} \beta_{s_{ij}} \rangle\right)$

$$= \left(\langle \sqrt[3]{\alpha_{s_{ij}}{}^3 + \alpha_{r_{ij}}{}^3 - \alpha_{s_{ij}}{}^3 \alpha_{r_{ij}}{}^3}, \beta_{s_{ij}} \beta_{r_{ij}} \rangle\right)$$

$$= S \oplus_F R.$$

To Prove (b):

$$(R \odot_F S) = \left(\langle \alpha_{r_{ij}} \alpha_{s_{ij}}, \sqrt[3]{\beta_{r_{ij}}{}^3 + \beta_{s_{ij}}{}^3 - \beta_{r_{ij}}{}^3 \beta_{s_{ij}}{}^3} \rangle\right)$$

$$= \left(\langle \alpha_{s_{ij}} \alpha_{r_{ij}}, \sqrt[3]{\beta_{s_{ij}}{}^3 + \beta_{r_{ij}}{}^3 - \beta_{s_{ij}}{}^3 \beta_{r_{ij}}{}^3} \rangle\right)$$

$$= S \odot_F R.$$

To Prove (c): $(R \oplus_F S)^c = \left(\langle \beta_{r_{ij}} \beta_{s_{ij}}, \sqrt[3]{\alpha_{r_{ij}}{}^3 + \alpha_{s_{ij}}{}^3 - \alpha_{r_{ij}}{}^3 \alpha_{s_{ij}}{}^3} \rangle\right)$

$$= R^c \odot_F S^c$$

To Prove (d): $(R \odot_F S)^c = \left(\langle \sqrt[3]{\beta_{r_{ij}}{}^3 + \beta_{s_{ij}}{}^3 - \beta_{r_{ij}}{}^3 \beta_{s_{ij}}{}^3}, \alpha_{r_{ij}} \alpha_{s_{ij}} \rangle\right)$

$$= R^c \oplus_F S^c$$

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Which shows FFMs satisfies commutativity and D'Morgan's laws.

Theorem 3.14:

Let T be FFM and 0 be the identity FFM with respect to $\oplus_F$ and J=<1, 0> be the identity $\odot_F$ then

(a) $T \oplus_F 0 = 0 \oplus_F T = T$

(b) $T\odot_F J = J \odot_F T = T$

Proof:

$$T = (< \alpha_{t_{ij}}, \beta_{t_{ij}} >)$$

$$T \oplus_F 0 = \left(\langle \sqrt[3]{\alpha_{t_{ij}}^3 + 0 - 0}, \beta_{t_{ij}}\rangle\right)$$

$$= \left(\langle \alpha_{t_{ij}}, \beta_{t_{ij}}\rangle\right) = T$$

$$T\odot_F J = \left(< \alpha_{t_{ij}}, \beta_{t_{ij}} >\right)\odot_F (< 1,0 >)$$

$$= \left(\langle \alpha_{t_{ij}}.1, \sqrt[3]{\beta_{t_{ij}}^3 + 0 - \beta_{t_{ij}}^3.0}\rangle\right)$$

$$= \left(\langle \alpha_{t_{ij}}, \beta_{t_{ij}}\rangle\right) = T$$

Similarly, $0 \oplus_F T = T$ and $J\odot_F T$= T can be proved.

Theorem 3.15: Let $R, S, T \in \mathcal{F}_{mn}$ then

(a) $(T \oplus_F R) \oplus_F S = T \oplus_F (R \oplus_F S)$

(b) $(T\odot_F R)\odot_F S = T\odot_F (R\odot_F S)$

Proof:

To Prove(a):Consider

$$(R \oplus_F S) = \left(\langle \sqrt[3]{\alpha_{r_{ij}}^3 + \alpha_{s_{ij}}^3 - \alpha_{r_{ij}}^3\alpha_{s_{ij}}^3}, \beta_{r_{ij}}\beta_{s_{ij}}\rangle\right)$$

$$T \oplus_F (R \oplus_F S) = \left(\langle \alpha_{t_{ij}}, \beta_{t_{ij}}\rangle\right) \oplus_F \left(\langle \sqrt[3]{\alpha_{r_{ij}}^3 + \alpha_{s_{ij}}^3 - \alpha_{r_{ij}}^3\alpha_{s_{ij}}^3}, \beta_{r_{ij}}\beta_{s_{ij}}\rangle\right)$$

$$= \left(\langle \sqrt[3]{\alpha_{r_{ij}}^3 + \alpha_{s_{ij}}^3 - \alpha_{r_{ij}}^3\alpha_{s_{ij}}^3 + \alpha_{t_{ij}}^3 - \alpha_{r_{ij}}^3\alpha_{t_{ij}}^3 - \alpha_{t_{ij}}^3\alpha_{s_{ij}}^3 - \alpha_{r_{ij}}^3\alpha_{s_{ij}}^3\alpha_{t_{ij}}^3}, \beta_{r_{ij}}\beta_{s_{ij}}\beta_{t_{ij}}\rangle\right)$$

$$= \left(\langle \sqrt[3]{\alpha_{r_{ij}}^3 + \alpha_{s_{ij}}^3 + \alpha_{t_{ij}}^3 - \alpha_{r_{ij}}^3\alpha_{s_{ij}}^3 - \alpha_{r_{ij}}^3\alpha_{t_{ij}}^3 - \alpha_{t_{ij}}^3\alpha_{s_{ij}}^3 - \alpha_{r_{ij}}^3\alpha_{s_{ij}}^3\alpha_{t_{ij}}^3}, \beta_{r_{ij}}\beta_{s_{ij}}\beta_{t_{ij}}\rangle\right) \ldots\ldots\ldots(1)$$

Consider the LHS

$$(T \oplus_F R) \oplus_F S = \left(\langle \sqrt[3]{\alpha_{t_{ij}}^3 + \alpha_{r_{ij}}^3 - \alpha_{r_{ij}}^3\alpha_{t_{ij}}^3}, \beta_{r_{ij}}\beta_{t_{ij}}\rangle\right) \oplus_F (< \alpha_{s_{ij}}, \beta_{s_{ij}} >)$$

$$= \left(\langle \sqrt[3]{\alpha_{r_{ij}}^3 + \alpha_{s_{ij}}^3 + \alpha_{t_{ij}}^3 - \alpha_{r_{ij}}^3\alpha_{s_{ij}}^3 - \alpha_{r_{ij}}^3\alpha_{t_{ij}}^3 - \alpha_{t_{ij}}^3\alpha_{s_{ij}}^3 - \alpha_{r_{ij}}^3\alpha_{s_{ij}}^3\alpha_{t_{ij}}^3}, \beta_{r_{ij}}\beta_{s_{ij}}\beta_{t_{ij}}\rangle\right)\ldots(2)$$

From (1)&(2) (a) is proved

To Prove (b):

RHS=$(T\odot_F R)\odot_F S = \left(\langle \alpha_{r_{ij}}\alpha_{t_{ij}}, \sqrt[3]{\beta_{r_{ij}}^3 + \beta_{t_{ij}}^3 - \beta_{r_{ij}}^3\beta_{t_{ij}}^3}\rangle\right) \odot_F \left(\langle \alpha_{s_{ij}}, \beta_{s_{ij}}\rangle\right)$

$$= \left(\langle \alpha_{r_{ij}}\alpha_{s_{ij}}\alpha_{t_{ij}}, \sqrt[3]{\beta_{r_{ij}}^3 + \beta_{s_{ij}}^3 - \beta_{r_{ij}}^3\beta_{s_{ij}}^3 + \beta_{t_{ij}}^3 - \beta_{r_{ij}}^3\beta_{t_{ij}}^3 - \beta_{t_{ij}}^3\beta_{s_{ij}}^3 - \beta_{r_{ij}}^3\beta_{s_{ij}}^3\beta_{t_{ij}}^3}\rangle\right)\ldots\ldots(3)$$

LHS=$T\odot_F (R\odot_F S) = \left(\langle \alpha_{t_{ij}}, \beta_{t_{ij}}\rangle\right) \odot_F \left(\langle \alpha_{s_{ij}}\alpha_{r_{ij}}, \sqrt[3]{\beta_{s_{ij}}^3 + \beta_{r_{ij}}^3 - \beta_{s_{ij}}^3\beta_{r_{ij}}^3}\rangle\right)$

$$= \left(\langle \alpha_{r_{ij}}\alpha_{s_{ij}}\alpha_{t_{ij}}, \sqrt[3]{\beta_{r_{ij}}^3 + \beta_{s_{ij}}^3 - \beta_{r_{ij}}^3\beta_{s_{ij}}^3 + \beta_{t_{ij}}^3 - \beta_{r_{ij}}^3\beta_{t_{ij}}^3 - \beta_{t_{ij}}^3\beta_{s_{ij}}^3 - \beta_{r_{ij}}^3\beta_{s_{ij}}^3\beta_{t_{ij}}^3}\rangle\right)\ldots\ldots(4$$

Hence $(T\odot_F R)\odot_F S = T\odot_F (R\odot_F S)$

Theorem 3.16:

Let $T, S, R \in \mathcal{F}_{mn}$ then

(a) $(T \vee R) \oplus_F S = (T \oplus_F S) \vee (R \oplus_F S)$

(b) $(T \wedge R) \oplus_F S = (T \oplus_F S) \wedge (R \oplus_F S)$

(c) $T\oplus_F (R \vee S) = (T \oplus_F R) \vee (T \oplus_F S)$

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(d) $T \oplus_F (R \wedge S) = (T \oplus_F R) \wedge (T \oplus_F S)$

Proof:To prove (a)

LHS=$\left(\langle max\left(\alpha_{r_{ij}}, \alpha_{t_{ij}} \right), min\left(\beta_{r_{ij}}, \beta_{t_{ij}} \right) \rangle \right) \oplus_F \left(\langle \alpha_{s_{ij}}, \beta_{s_{ij}} \rangle \right)$

$= \left(\langle max\left(\sqrt[3]{\alpha_{r_{ij}}^3 + \alpha_{s_{ij}}^3 - \alpha_{r_{ij}}^3 \alpha_{s_{ij}}^3}, \sqrt[3]{\alpha_{s_{ij}}^3 + \alpha_{t_{ij}}^3 - \alpha_{s_{ij}}^3 \alpha_{t_{ij}}^3} \right), min\left(\beta_{r_{ij}} \beta_{s_{ij}}, \beta_{s_{ij}} \beta_{t_{ij}} \right) \rangle \right)$

$= \left(\langle \sqrt[3]{\alpha_{r_{ij}}^3 + \alpha_{s_{ij}}^3 - \alpha_{r_{ij}}^3 \alpha_{s_{ij}}^3}, \beta_{r_{ij}} \beta_{s_{ij}} \rangle \right) \vee \left(\langle \sqrt[3]{\alpha_{s_{ij}}^3 + \alpha_{t_{ij}}^3 - \alpha_{s_{ij}}^3 \alpha_{t_{ij}}^3}, \beta_{s_{ij}} \beta_{t_{ij}} \rangle \right)$

$= (R \oplus_F S) \vee (T \oplus_F S)$

=RHS

To Prove (b):

LHS= $\left(\langle min(\alpha_{r_{ij}}, \alpha_{t_{ij}}), max(\beta_{r_{ij}}, \beta_{t_{ij}}) \rangle \right) \oplus_F \left(\langle \alpha_{s_{ij}}, \beta_{s_{ij}} \rangle \right)$

$= \left(\langle min\left(\sqrt[3]{\alpha_{r_{ij}}^3 + \alpha_{s_{ij}}^3 - \alpha_{r_{ij}}^3 \alpha_{s_{ij}}^3}, \sqrt[3]{\alpha_{s_{ij}}^3 + \alpha_{t_{ij}}^3 - \alpha_{s_{ij}}^3 \alpha_{t_{ij}}^3} \right), max\left(\beta_{r_{ij}} \beta_{s_{ij}}, \beta_{s_{ij}} \beta_{t_{ij}} \right) \rangle \right)$

$= \left(\langle \sqrt[3]{\alpha_{r_{ij}}^3 + \alpha_{s_{ij}}^3 - \alpha_{r_{ij}}^3 \alpha_{s_{ij}}^3}, \beta_{r_{ij}} \beta_{s_{ij}} \rangle \right) \wedge \left(\langle \sqrt[3]{\alpha_{s_{ij}}^3 + \alpha_{t_{ij}}^3 - \alpha_{s_{ij}}^3 \alpha_{t_{ij}}^3}, \beta_{s_{ij}} \beta_{t_{ij}} \rangle \right)$

$= (R \oplus_F S) \wedge (T \oplus_F S)$

=RHS.

To Prove (c):

LHS= $\left(\langle \alpha_{t_{ij}}, \beta_{t_{ij}} \rangle \right) \oplus_F \left(\langle max\left(\alpha_{s_{ij}}, \alpha_{r_{ij}} \right), min\left((\beta_{s_{ij}}, \beta_{r_{ij}}) \right) \rangle \right)$

$= \left(\langle max\left(\sqrt[3]{\alpha_{t_{ij}}^3 + \alpha_{s_{ij}}^3 - \alpha_{t_{ij}}^3 \alpha_{s_{ij}}^3}, \sqrt[3]{\alpha_{r_{ij}}^3 + \alpha_{t_{ij}}^3 - \alpha_{r_{ij}}^3 \alpha_{t_{ij}}^3} \right), min\left((\beta_{t_{ij}} \beta_{s_{ij}}, \beta_{t_{ij}} \beta_{r_{ij}}) \right) \rangle \right)$

$= \left(\langle \sqrt[3]{\alpha_{t_{ij}}^3 + \alpha_{s_{ij}}^3 - \alpha_{t_{ij}}^3 \alpha_{s_{ij}}^3}, \beta_{t_{ij}} \beta_{s_{ij}} \rangle \right) \vee \left(\langle \sqrt[3]{\alpha_{r_{ij}}^3 + \alpha_{t_{ij}}^3 - \alpha_{r_{ij}}^3 \alpha_{t_{ij}}^3}, \beta_{r_{ij}} \beta_{t_{ij}} \rangle \right)$

$= (T \oplus_F S) \vee (T \oplus_F R)$

To Prove (d):

LHS=$\left(\langle \alpha_{t_{ij}}, \beta_{t_{ij}} \rangle \right) \oplus_F \left(\langle min\left(\alpha_{s_{ij}}, \alpha_{r_{ij}} \right), max\left(\beta_{s_{ij}}, \beta_{r_{ij}} \right) \rangle \right)$

$= \left(\langle min\left(\sqrt[3]{\alpha_{t_{ij}}^3 + \alpha_{s_{ij}}^3 - \alpha_{t_{ij}}^3 \alpha_{s_{ij}}^3}, \sqrt[3]{\alpha_{r_{ij}}^3 + \alpha_{t_{ij}}^3 - \alpha_{r_{ij}}^3 \alpha_{t_{ij}}^3} \right), max\left(\beta_{t_{ij}} \beta_{s_{ij}}, \beta_{r_{ij}} \beta_{t_{ij}} \right) \rangle \right)$

$= \left(\langle \sqrt[3]{\alpha_{t_{ij}}^3 + \alpha_{s_{ij}}^3 - \alpha_{t_{ij}}^3 \alpha_{s_{ij}}^3}, \beta_{t_{ij}} \beta_{s_{ij}} \rangle \right) \wedge \left(\langle \sqrt[3]{\alpha_{r_{ij}}^3 + \alpha_{t_{ij}}^3 - \alpha_{r_{ij}}^3 \alpha_{t_{ij}}^3}, \beta_{r_{ij}} \beta_{t_{ij}} \rangle \right)$

$= (T \oplus_F S) \wedge (T \oplus_F R)$

=RHS.

Theorem 3.17:

Let $R, S, T \in \mathcal{F}_{mn}$ then (a) $(R \vee S) \odot_F T = (R \odot_F T) \vee (S \odot_F T)$

(b) $(R \wedge S) \odot_F T = (R \odot_F T) \wedge (S \odot_F T)$

(c) $R \odot_F (S \vee T) = (R \odot_F S) \vee (R \odot_F T)$

(d) $R \odot_F (S \wedge T) = (R \odot_F S) \wedge (R \odot_F T)$

Proof: To Prove (a):

LHS=$(R \vee S) \odot_F T = \left(\langle max\left(\alpha_{r_{ij}}, \alpha_{s_{ij}} \right), min\left(\beta_{r_{ij}}, \beta_{s_{ij}} \right) \rangle \right) \odot_F \left(\langle \alpha_{t_{ij}}, \beta_{t_{ij}} \rangle \right)$

$= \left(\langle max\left(\alpha_{r_{ij}} \alpha_{t_{ij}}, \alpha_{s_{ij}} \alpha_{t_{ij}} \right), min\left(\sqrt[3]{\beta_{r_{ij}}^3 + \beta_{t_{ij}}^3 - \beta_{r_{ij}}^3 \beta_{t_{ij}}^3}, \sqrt[3]{\beta_{s_{ij}}^3 + \beta_{t_{ij}}^3 - \beta_{s_{ij}}^3 \beta_{t_{ij}}^3} \right) \rangle \right)$

$= \left(\langle \alpha_{r_{ij}} \alpha_{t_{ij}}, \sqrt[3]{\beta_{r_{ij}}^3 + \beta_{t_{ij}}^3 - \beta_{r_{ij}}^3 \beta_{t_{ij}}^3} \rangle \right) \vee \left(\langle \alpha_{s_{ij}} \alpha_{t_{ij}}, \sqrt[3]{\beta_{s_{ij}}^3 + \beta_{t_{ij}}^3 - \beta_{s_{ij}}^3 \beta_{t_{ij}}^3} \rangle \right)$

$= (R \odot_F T) \vee (S \odot_F T)$=RHS

To Prove (b):

LHS=$(R \wedge S) \odot_F T = \left(\langle min\left((\alpha_{r_{ij}}, \alpha_{s_{ij}}), max\left(\beta_{r_{ij}}, \beta_{s_{ij}} \right) \right) \rangle \right) \odot_F \left(\langle \alpha_{t_{ij}}, \beta_{t_{ij}} \rangle \right)$

$= \left(\langle min\left(\alpha_{r_{ij}} \alpha_{t_{ij}}, \alpha_{s_{ij}} \alpha_{t_{ij}} \right), max\left(\sqrt[3]{\beta_{r_{ij}}^3 + \beta_{t_{ij}}^3 - \beta_{r_{ij}}^3 \beta_{t_{ij}}^3}, \sqrt[3]{\beta_{s_{ij}}^3 + \beta_{t_{ij}}^3 - \beta_{s_{ij}}^3 \beta_{t_{ij}}^3} \right) \rangle \right)$

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$$= \left(\langle \alpha_{r_{ij}} \alpha_{t_{ij}}, \sqrt[3]{\beta_{r_{ij}}{}^3 + \beta_{t_{ij}}{}^3 - \beta_{r_{ij}}{}^3 \beta_{t_{ij}}{}^3} \rangle \right) \wedge \left(\langle \alpha_{s_{ij}} \alpha_{t_{ij}}, \sqrt[3]{\beta_{s_{ij}}{}^3 + \beta_{t_{ij}}{}^3 - \beta_{s_{ij}}{}^3 \beta_{t_{ij}}{}^3} \rangle \right)$$

$= (R \odot_F T) \wedge (S \odot_F T)$

=RHS

To Prove (c):

$\text{LHS} = R \odot_F (S \vee T) = \left(\langle \alpha_{r_{ij}}, \beta_{r_{ij}} \rangle \right) \odot_F \left(\langle max\left(\alpha_{s_{ij}}, \alpha_{t_{ij}} \right), min\left(\beta_{s_{ij}}, \beta_{t_{ij}} \right) \rangle \right)$

$$= \left(\langle max\left(\alpha_{r_{ij}} \alpha_{s_{ij}}, \alpha_{r_{ij}} \alpha_{t_{ij}} \right), min\left(\sqrt[3]{\beta_{r_{ij}}{}^3 + \beta_{s_{ij}}{}^3 - \beta_{r_{ij}}{}^3 \beta_{s_{ij}}{}^3}, \sqrt[3]{\beta_{r_{ij}}{}^3 + \beta_{t_{ij}}{}^3 - \beta_{r_{ij}}{}^3 \beta_{t_{ij}}{}^3} \right) \rangle \right)$$

$$= \left(\langle \alpha_{r_{ij}} \alpha_{s_{ij}}, \sqrt[3]{\beta_{r_{ij}}{}^3 + \beta_{s_{ij}}{}^3 - \beta_{r_{ij}}{}^3 \beta_{s_{ij}}{}^3} \rangle \right) \vee \left(\langle \alpha_{r_{ij}} \alpha_{t_{ij}}, \sqrt[3]{\beta_{r_{ij}}{}^3 + \beta_{t_{ij}}{}^3 - \beta_{r_{ij}}{}^3 \beta_{t_{ij}}{}^3} \rangle \right)$$

$= (R \odot_F S) \vee (R \odot_F T)$

=RHS.

To Prove (d):

Consider the LHS

$R \odot_F (S \wedge T) = \left(\alpha_{r_{ij}}, \beta_{r_{ij}} \right) \odot_F \left(min\left(\alpha_{s_{ij}}, \alpha_{t_{ij}} \right), max\left(\beta_{s_{ij}}, \beta_{t_{ij}} \right) \right)$

$$= \left(min\left(\alpha_{r_{ij}} \alpha_{s_{ij}}, \alpha_{r_{ij}} \alpha_{t_{ij}} \right), max\left(\sqrt[3]{\beta_{r_{ij}}{}^3 + \beta_{s_{ij}}{}^3 - \beta_{r_{ij}}{}^3 \beta_{s_{ij}}{}^3}, \sqrt[3]{\beta_{r_{ij}}{}^3 + \beta_{t_{ij}}{}^3 - \beta_{r_{ij}}{}^3 \beta_{t_{ij}}{}^3} \right) \right)$$

$$= \left(\alpha_{r_{ij}} \alpha_{s_{ij}}, \sqrt[3]{\beta_{r_{ij}}{}^3 + \beta_{s_{ij}}{}^3 - \beta_{r_{ij}}{}^3 \beta_{s_{ij}}{}^3} \right) \wedge \left(\alpha_{r_{ij}} \alpha_{t_{ij}}, \sqrt[3]{\beta_{r_{ij}}{}^3 + \beta_{t_{ij}}{}^3 - \beta_{r_{ij}}{}^3 \beta_{t_{ij}}{}^3} \right)$$

$= (R \odot_F S) \wedge (R \odot_F T)$

=RHS.

Theorem 3.18:

Let $R, S \in \mathcal{F}_{mn}$ with $S \leq R$ then

(a) $(S \wedge R) \oplus_F (S \vee R) = (S \oplus_F R)$

(b) $(S \wedge R) \odot_F (S \vee R) = (S \odot_F R)$

Proof: To Prove (a):$= (S \wedge R) \oplus_F (S \vee R)$

$= \left(\langle min\left(\alpha_{r_{ij}}, \alpha_{s_{ij}} \right), max\left(\beta_{r_{ij}}, \beta_{s_{ij}} \right) \rangle \right) \oplus_F \left(\langle max\left(\alpha_{r_{ij}}, \alpha_{s_{ij}} \right), min\left(\beta_{r_{ij}}, \beta_{s_{ij}} \right) \rangle \right)$

$= \left(\langle \alpha_{s_{ij}}, \beta_{s_{ij}} \rangle \right) \oplus_F \left(\langle \alpha_{r_{ij}}, \beta_{r_{ij}} \rangle \right) = S \oplus_F R$

To Prove (b):

$= (S \wedge R) \odot_F (S \vee R)$

$= \left(\langle min\left(\alpha_{r_{ij}}, \alpha_{s_{ij}} \right), max\left(\beta_{r_{ij}}, \beta_{s_{ij}} \right) \rangle \right) \odot_F \left(m \langle ax\left(\alpha_{r_{ij}}, \alpha_{s_{ij}} \right), min\left(\beta_{r_{ij}}, \beta_{s_{ij}} \right) \rangle \right)$

$= \left(\langle \alpha_{s_{ij}}, \beta_{s_{ij}} \rangle \right) \odot_F \left(\langle \alpha_{r_{ij}}, \beta_{r_{ij}} \rangle \right)$

$= (S \odot_F R).$

CONCLUSION

The concepts of Fermatean Fuzzy Matrix, addition, and multiplication operators have been introduced in this study and forms a commutative monoid. We've also talked about conjuction and disjunction to talk about some of their features. On fermatean fuzzy matrices, we also formulate and show some inequalities related to algebraic sum and algebraic product operations.

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REVIEW ARTICLE

A Comprehensive Review on the Reinforced Concrete Walls with Openings

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ABSTRACT

In recent decades, R.C. walls are the most appropriate structural forms, which have caused the height of concrete buildings to be soared. Due to the role they play as a structure's horizontal force resisting system, the ability to accurately predict and model shear wall behavior is of the utmost importance. R.C walls in buildings might have openings with different configurations of openings due to architectural purposes. The size, position, and area of openings may change from an engineering perspective. In this review studied experimental output including load capacity, failure mode, as well as crack pattern and lateral displacement. A parametric study is conducted to investigate the effect of opening size, shape, orientation and aspect ratio in R.C. wall. In addition, another parametric study is conducted to investigate the effect of pre and post-strengthening schemes in the one-way R.C. wall with opening. Also, the effect of increasing the horizontal and vertical externally bonded Carbon Fiber Reinforced Polymer (CFRP) sheet development length wall is studied (all around the opening 90o). Moreover, the influence of geometric characteristics, re-arrangement and dividing the vertical CFRP sheets on increasing the axial capacity of R.C. walls is examined. The results demonstrated that opening shape, orientation, aspect ratio and area should be taken into consideration as they affect the axial capacity of the wall. It is also recommended to increase CFRP development length paralleled to the load direction to about 22% of the opening height. It is also concluded that the most effective strengthening scheme to increase the axial load capacity is using with one wide externally bonded CFRP sheet instead of re-arranging it to equal spaced strips which have the same total sheet area.

Key words: CFRP, R.C. Walls, Opening, Finite Element Analysis

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INTRODUCTION

An earthquake is a sudden motion or shaking of the Earth's crust, caused by the abrupt release of slowly accumulated strain stored in the rocks beneath the surface [1, 2], it generates elastic vibration or waves which movement all direction from the point of origin and cause earthquake.

There are four basic causes of earthquake: -
(i) Ground shaking (ii) ground failure (iii) tsunamis (iv) fire.
In tall buildings lateral loads are premier one which will increase rapidly with increase in height. The design takes care of the requirements of strength, rigidity and stability. The structural system designed to carry vertical load may not have the capacity to resist lateral load or even if it has, the design for lateral load will increase the structural cost substantially with increase in number of story [3, 4].

Shear wall structures: A shear wall is a structural system providing stability against wind, earthquake and blast deriving its stiffness from inherent structural forms .

There are different types of shear walls such as given below:-
- Cantilever shear walls
- Flanged cantilever shear walls
- Coupled shear walls
- Shear wall with openings
- Box system
-

Among different systems in structures and components, using RC walls in construction is becoming increasingly popular. The fast installation, time-saving and the possibility of prefabrication, make RC walls potentially one of the most desirable construction systems. They are efficient, both in terms of construction cost and effectiveness in minimizing earthquake damage in structural and non-structural elements [5, 6]. Although the overall budget for the structure increases slightly, the enhanced structural efficiency balances out these costs. Steel reinforcing bars are to be placed in walls in regularly spaced vertical and horizontal grids. The vertical and horizontal reinforcement in the wall can be placed in one or two parallel layers called curtains [7].

Behavior of shear walls: Several methods of shear walls have been described by number of authors. The actual behavior of shear wall under earthquake loading is inelastic but for simplification of computations its behavior is assumed elastic in most of the methods [8]. On basis of its behavior analysis is divided into two major categories. i.e. linear and nonlinear. The most commonly used methods in linear category are continuum approach, equivalent wide column modeling, and equivalent shear wall-frame system analysis and finite element method. The most logical approach to the seismic design problem is to accept the uncertainty of the seismic phenomenon and consequently to design the structure in such a way that an adequate reserve of resistance is available to prevent failure in the case of a major earthquake, but little or no additional cost compared to designing the structure to resist frequent earthquake motions [9, 10].In order to achieve safe, economic and functional buildings, strengthening of structural members using several methods has gained a great deal of attention in recent years and proved the efficiency in increasing the capacity of the R.C. walls with opening.

The openings are source of weak points and cause decrease inside the structure's load-bearing capacity and stiffness .Strengthening R.C. wall is divided into two main types: pre-strengthening and post-strengthening. Pre-strengthening includes embedding extra internal steel bars around openings before concrete casting. On the other hand, Post strengthening includes using externally bonded Fiber Reinforced Polymer (FRP) sheets with different development lengths and Near Surface Mounted (N.S.M) FRP reinforcement and steel bars during the R.C. wall

service life. Based on the loading system, different types of failures may result. The in plane loading may induce diagonal compressive failure, diagonal tensile failure, or concrete crushing due to bending, while the perpendicular loading produces an out of plane bending failure. In the case of gravitational load, the most common failure is compressive failure if the load is not eccentric. In some cases, more than one mode of failure can occur in RC walls when it is under several simultaneous loadings. The presence of an opening in RC wall under eccentric axial loads results in local cracking in the vicinity of the opening as well as a reduction in load carrying capacity. Therefore, this necessitates an improvement of the ultimate load bearing capacity by exploring the best opening configuration and strengthening RC walls [11].The following sections discuss the effect of openings in R.C. walls and previous proposed formulae to determine the ultimate load of RC walls with openings. Then, an overview of experimental programs undertaken by previous researchers on strengthened RC walls was conducted

Eccentric Openings

Rafik Taleb et al., [76] ,The main purposes of the experimental tests were to evaluate the shear behavior and to clarify the influence of opening ratios on the cracks distribution and the shear strength of structural walls under horizontal reversed cyclic loading. Tested Four Reinforced Concrete (RC) single span structural walls having various opening sizes and locations under lateral reversed cyclic loading ,These specimens were scaled to 40% and represented the lower three stories of a six storied RC building. The specimens were three-storied and 40% scaled models. As shown in Figure 43, three of these specimens (S1, M1, L1) were with eccentric openings and one specimen (N1) without openings. For specimens with openings, the main test variables were the opening ratio and the opening location. The opening ratios were 0.3, 0.34 and 0.46 for S1, M1 and L1 specimen, respectively. All the specimens were 4150 mm height and 2800 mm wide. The beams were nominally 200 mm wide by 300 mm deep and the side columns were 300 mm by 300 mm. The thickness of wall panels was 80 mm. To provide a fixed base at the bottom, a RC foundation beam with 600 mm wide by 400 mm thick and 3600 mm in length was built integrally with the body of the structural walls and post-tensioned to the reaction floor prior to testing. The clear span was 2200 mm and the column clear story height was 1100 mm, 1100 mm and 550 mm in the first, second and third story, respectively. A 400 mm wide by 400 mm deep loading beam was cast at the top of the wall panel. A hydraulic actuator was attached to the specimen at mid-span of the loading beam to apply the horizontal reversed cyclic loading. The third story was provided for releasing the confinement caused by the stiff loading beam at the top. The structural walls were tested in a lateral reverse cyclic manner until their maximum performances. Since one of the purposes of this study is to clarify the influence of the opening ratios on the shear behaviour, all the specimens were designed to fail in shear and not in flexure

The lateral load, Q, was applied statically to the loading beam by two 2 MN hydraulic jacks. Cyclic reversed horizontal loads were statically applied to the specimens in both positive and negative directions. During the cyclic horizontal loading, vertical axial loads were applied to columns by two 1 MN hydraulic jacks assuming that the specimens are representing the lower three stories of a typical six stories RC building. The vertical axial load levels were determined in accordance with the assumed long-term axial loads for a six-story wall with three spans.

The lateral reinforcing bars of the first story wall panel were bent severely, and cracks have propagated extensively along the wall reinforcements. For this specimen, L1, the strength degradation after the peak load did not drop suddenly like observed for the case of Specimens S1 and M1. Specimen S1 failed by shear failure of the short span beams, while M1 failed by shear sliding at the first story wall panel. On the other hand, L1 failed in a ductile manner after flexural yielding of the short span beam took place followed by shear sliding of the wall panel at the final stage.

For all specimens, main reinforcements of side columns at the bottom of the first story yielded at the drift angle of about 1.0%, while those of short span beams yielded at the drift angle of about 0.25%. All stirrups of the short span beams of S1 and M1 yielded, while those of short span beam of L1 yielded at the first story only. Most of the lateral reinforcements of wall panel yielded around the peak load corresponding to the drift angle of about 0.5%. For walls with openings, most of the reinforcing bars around the openings were yielded at the drift angle of about 1.0%. RafikTaleb et al., [76] drew a number of conclusions, some of which are outlined as follows.

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Due to the eccentric opening location Shear strength of a structural wall was different between positive and negative loading directions. Shear strength obtained while loading from the opening side was larger than that obtained from the opposite side. The raison was due to the existence of eccentric openings that affected the formation of concrete strut. Shear strength of small opening ratio was higher than those of with larger opening ratio. However, Opening ratios affect the shear strength especially when the openings of the structural walls are at the same location. It is worth to mention that, damage to the compressed concrete of the first story wall, was the principal factor which influenced the shear capacity of the multi-story structural walls.

Impact of Openings on R.C. Wall Behavior under Vertical Load Axial Capacity

Bashar Mohammed et al.,[23] tested axially sixteen 1/3 scale reinforced concrete wall panels with openings comprising four identical series of four specimens. All the test panels are 400mm in width and 800mm in height. Series one designated WO1-WO4, are 50mm thick and 3 other series were 40mm in thickness. Series one and two designated as WO1-WO4 and WO1a-WO4a, respectively are tested without CFRP sheet; while series three and four designated as WO1b-WO4b and WO1c-WO4c,respectively are tested with CFRP sheet strengthened wall openings. Wall panels are tested with different sizes of openings. Opening sizes increase by percentage of wall area of 5% (WO1: 95 mm x 170mm, 10% (WO2: 135 mm x 240 mm), 20% (WO3: 185mm x 340 mm) and 30% (WO4: 230 mm x 420 mm). Aspect ratio (H/L), slenderness ratio (H/t) and thinness ratio (L/t) are 2, 16 and 8 respectively for wall series one. For series two, three, and four, aspect ratio (H/L), slenderness ratio (H/t) and thinness ratio(L/t) are 2, 20 and 10, respectively. The specimens are loaded by axial loads and reinforced by centrally positioned single layer of welded wire fabric reinforcement, consisting of deformed 5 mm diameter bars. There are hinged connections at the top and bottom boundaries of the specimen and free side edges. Steel mean yield strength (fy) was 478 MPa. Regarding the strengthened specimens, CFRP sheets are externally bonded alongside the opening with thickness (0.167 mm) and tensile modulus 230000 Mpa. A hydraulic jack with a with a most extreme limit of 300 kN, applies a distributed uniform load, with controlled total force, along the wall length. The specimens were subjected to an eccentricity of one sixth of the wall thickness and a steel rod was welded to both of loading beam in order to apply eccentric distributed loading, designed to fit into a guide system connected to the upper edge and lower edge of the specimen as illustrated in figure 5[23].

This study showed that there is a decrease in the theoretical failure loads (Pcuo) & experimental axial capacity (Peuo) when the opening size increases. Devika Sagar V. et al.,[31] carried out experimental investigation to find the effect of different types of openings on the strength and behavior of R.C. wall panels under one way in-plane action. Six numbers of square wall panels of size 750 mm and 35 mm thickness were cast and tested axially. Size of window opening provided was 300 x 375mm and that of door opening was 300 x 525 mm. As illustrated in figure 6, it is seen that the first crack load is found to decrease by 17% and the ultimate load by 21% for20 % reduction in area due to window opening. The reduction in strength is 44% and47% respectively for first crack load and ultimate load corresponding 28% reduction in area in case of panels with door opening. Nh at-Minh Ho et al.,[42] on 12 half scale walls, including experimental setup, failure loads, crack patterns, and load deflection characteristics, are reported. Typical details of the test panels are given in figure 7. In addition, the Finite Element Method using ABAQUS software for investigating the behavior of TW3S (two way three sided) walls is described in detail in figure 8. In the experimental program, 12 half-scale RC wall panels, including four solid panels and eight panels with an opening, were constructed and tested to failure. The slenderness ratios of these walls varied from 25 to 40 along with varying aspect ratios from 0.83 to 1.60. The ratio of opening size to total area of wall (Ao/Aw, where Ao=Ho×Lo and Aw=Hw×Lw) was chosen to be 5% or 10%, where the height of the opening was equal to 1/3 of the height of the wall.

As illustrated in figure 8, based on the experimental results, the 5% and 10%reductions in the area of the solid wall (Hw/tw = 30, Hw/Lw = 1.2) caused by introducing the openings at the center reduced its load carrying capacity by 10.2% and17.0%,respectively. The FEM results reveal 18.7% and 32.0% reductions in wall strength, respectively.

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Ductility & Energy Absorption Capacity

Ductility is commonly expressed in terms of displacements, curvature or rotations. Displacement-based ductility factors (defined as the ratios between elastic and ultimate displacements, $\mu\Delta=du/de$) are computed. In addition to ductility factors‚energy dissipation (Ed) was also evaluated as the area under the load-displacement curves. Devika Sagar V. et al.,[31] whose experimental program revealed that Energy absorption capacity (the area under the load-deflection graph up to ultimate load) was found to decrease with increase in area of openings. There was about 60% decrease in energy absorption capacity for panels with window openings and 78% decrease in energy absorption capacity for panels with door openings as presented in figure 9.

Cosmin Popescu, et al., [26] carried out an experimental program which is consisting of three R.C. wall specimens designed to represent typical wall panels in residential buildings (1800 mm long, 1350 mm tall and 60 mm thick), modeled for testing to failure. One of them is a solid wall I-C and the others have symmetric openings I-L(900 mm x 1050 mm) & I-S (450 mm x 1050 mm).The specimens are designed to be load-bearing concrete walls that are loaded by vertical loads with no transverse loads between supports or lateral in-plane forces. Welded wire fabric reinforcement was used to reinforce the walls, consisting of deformed 5 mm diameter bars with 100 mm spacing in both orthogonal directions and centrally positioned in a single layer. The concrete utilized to cast the specimens was a self consolidating blend that could be poured without vibrating it. The average cubic compressive strength of the concrete was 62.8 MPa. Steel mean yield strength (fy) was632 MPa.

Four hydraulic jacks, each with a most extreme limit of 1400 kN, were connected together to apply a uniformly distributed load, with controlled total force, along the wall length. An eccentricity of one sixth of the wall thickness was applied in the loading. A steel rod was welded to both of loading beam (top and bottom) in order to apply eccentric distributed loading, designed to fit into a guide system connected to the upper edge and lower edge of the specimen. This study showed that the introduction of the small and large openings resulted in similar, sharp reductions in computed ductility factors . However, the differences in size between the openings strongly affected the energy dissipation; the walls with no opening and a small opening could both be classified as "ductile elements" according to Park (1988), having ductility factors between 3 and 6 ‚while the wall with a large opening would be classified as an element with "restricted" ductility(ductility factor < 3).

Crack Pattern & Mode of Failure

Seddon et al., [53] tested monolithic concrete walls similar to the type found in construction under both pure axial and eccentric axial loading to investigate failure mechanisms, crack propagation and deflection behavior. The tested panels had slenderness ratios (H/tw) ranging from 18 to 54, concrete cube strengths ranging from13.4 MPa to 34.5 MPa, and were all supported in one-way action. Furthermore Seddonet al., [53] conducted tests on walls with openings under both concentrated and uniformly distributed loads using full size and half size concrete wall panels. The symmetrical openings were approximately half the wall length, with windows near the top of the panels and doors extending to the base. All openings were of practical dimensions similar to those found in practice. In his investigation, Seddon et al., [53] explained the structural behavior of wall panels with the presence of openings by introducing the concept of beam and column elements. Through his studies, Seddon et al., [53] drew a number of conclusions, some of which are outlined as follows:

- The openings caused beam element behavior above and below the openings.
- The portions adjacent to openings in the direction of the load exhibited column element action.
- It was found that cracking of beam elements occurred at small proportions of the ultimate load but the panels ultimately failed through one of the column elements due to cracks extending to the corners of openings

Zielinski et al., [54] tested five full scale panels (including one panel with an opening) with variable reinforcement and under axial and eccentric loading to determine the bearing capacity of thin wall-ribbed reinforced concrete panels. The experimental program focused on deflections, crack appearance and propagation, the behavior at junctions of thin walls with ribs, and the ultimate capacity. The aim of the study was to investigate whether the

empirical formula as given in ACI-318-05 for solid walls were applicable for thin wall-ribbed panels Zielinski et al., [64]. Each of the five panels was 38 mm thick, measured 1220 x 2745 mm, and was reinforced with affixed arrangement of cross ribs. One panel had a hole cut in the form of a window; another was loaded with an eccentricity of 6.5 mm, while all other panels were concentrically loaded. Panels were supported at the top and bottom only, and concrete strength was approximately 35 MPa. It was clearly observed that for the panel with an opening the failure occurred at the window corner, where a concentration of stresses occurred due to the abrupt change in cross section.

J. H. Doh et al., [16] tested eight numbers of half-scale reinforced concrete wall panels with opening one and two rectangular openings. The wall panels were either supported at top and bottom (one-way action) or restrained also on the side edges (two way action). The symbols OW and TW indicate one and two-way action tests, respectively. The first digit following the symbols denotes the number of openings; the second digit denotes the slenderness ratio with 1 for H/tw = 30 and 2 for 40For one-way walls with openings as shown in figure 11, it was noted that horizontal crack patterns developed in the column sections only, which is logical since this is the area where the cross-section is reduced. For two-way wall panels, from figures 12to 13, biaxial curvature manner is observed. In addition, cracks were observed at the corners of the wall panels. However the panels with two openings in figure 13shows failure occurs horizontally through the middle column element as well as from outer edges, thus again illustrating that failure is critical within the column element. Also cracking is also initiated at the corners of the openings. This suggests that the interior corners act as stress concentrators and thus initiating failure.

Parameters Affecting R.C. Walls with Openings Behaviorunder Vertical Load
Boundary Conditions.

Dong-Jun Lee, [36] tested forty-seven concrete wall panels with openings in three stages. All wall panels produced were square type. The lengths and heights of the panels were either 1200, 1400 or 1600mm with a thickness of 40mm. Eighteen wall specimens were tested in one-way action for the first stage followed by seventeen specimens tested in two-way action for the second stage. In each stage there were six wall type configurations. Each wall configuration consisted of three slenderness ratios. of 30, 35 and 40 with either one or two openings. Finally, for the third stage twelve specimens consisting of seven wall configurations with varying opening size and location were also tested in both one- and two-way actions (10≤ H/tw ≤30). The opening sectional area ratio was 25%, except for the special types of panel openings such as wide window, door and small window tested for the third stage.

The wall panel nomenclature is given as follows.
O 90 W 2 C 1.6
A letter at the first column indicates restraint condition of the wall panels such that O or T denotes that the test panels are in one- and two-way action, respectively. Two digital numbers in the second column indicates the nominal concrete characteristic strength designated, which is different from the actual mean strength obtained from the concrete cylinder test results of the wall panels. Next is a letter that represents a Window or Door type opening configuration and the following number indicates the number of openings. The fifth column refers to the location of opening, such as at Centre, Left side, upper or smaller window located at the Bottom. The last number indicates the wall panel length. Comparisons of load versus maximum lateral deflection responses of wall panels with two openings (OW2C1.2 and TW2C1.2; see figure 13 (a&b)) and walls with wide openings (OWW1.2 and TWW1.2; see figure 16 (a&b)) are presented in figure14. The responses of walls with wide opening are highlighted by grey color symbol. The ultimate failure load of the wall panel with wide opening (OWW1.2) in one-way action was slightly less than that of its counterpart (as shown in figure 14 (a)), where as that of wall TWW1.2 in two-way action was more than that of its counterparts(see figure 14 (b)). Also for one-way walls the axial strength of O50W2C1.2 (0.079)is slightly higher than that of O65W1W1.2 (0.061). However, for two-way walls theaxial strength of T70W2C1.2 (0.178) is significantly weaker than T65W1W1.2 (0.252). To further investigate the observations of walls in one-way action, the crack pattern sat failure and deflection profiles of walls O50W2C1.2 and O65W1W1.2 tested are reproduced in figure 14.

Sine curves (dotted lines) were linked to the maximum deflections through each quarter points (top and bottom) from each edge. Note from deflection profiles the top and bottom dial gauge measurements of the wall with two windows were much less than these of wall with wide window . It can be concluded that the top and bottom edges of wall with two windows were not allowed to fully rotate, which may have contributed to its extra capacity compared to wall with wide window. Deflection profiles for the wall with wide opening O65W1W1.2 showed a closer trend to its ideal sine curve. It is evident therefore that the capacity of walls with two openings would be anticipated to be greater than that with a wide opening in one-way action due to the supporting column action. For panels in two-way action, crack patterns at failure for walls T70W2C1.2 andT65W1W1.2 are reproduced . As discussed earlier, the failure load of the wall with two openings T70W2C1.2 was weaker than the wall with a singlewide opening (identical opening area) T65W1W1.2. This is mainly due to one-way actioned column area (indicated by dotted circle in figure 15 (a)). The horizontal cracks indicate a typical one-way behavior occurring at the mid height of column area It is evident therefore that the capacity of walls with two openings would be anticipated to be less than that with a wide opening in two way action due to the localized column action.

Opening Position

Dong-Jun Lee, [40] whose experimental program made comparisons of load versus maximum lateral deflection responses of wall panels with three different opening locations; center W1C1.2 (see figure 16 (a)), left side only WL (see figure 16 (b)) and left side on the center line WU (see figure 16(c)) are presented in figure 18 in one- and two-way actions .The wall behaviors in these cases were observed to be quite similar in both one- and two-way actions. The failure loads of wall panels with asymmetric opening (WL and WU highlighted by grey into big symbols) were observed to be slightly less than those of wall panels with symmetric opening (W1C1.2) in both one- and two-way actions for similar concrete strengths. A decrease in axial strength ratio of walls with asymmetric openings located at the left side and upper side of 26% and 27% was found for one-way action and 19%and 18% for two-way action respectively compared to the ratios of normal strength concrete walls with opening. It can be concluded that asymmetric openings contributed to reduction in axial strength of wall panels compared to a symmetric opening

Through their studies, Nhat-Minh Ho et al., [42] whose experimental & F.E. program concluded that while varying the opening location left, center, or right, along the central horizontal axis of the walls, led to 14.6%, 17.3%, and22.5% reductions in wall strengths, respectively. The FEM results in figure 18.demonstrate 24.8%, 30.9%, and 28.1% reductions in wall strength, respectively. The experimental and FEM results also highlight that the positions of the openings greatly affect the behavior of a TW3S wall as they appear to have offset the perceived advantages gained from having a side restraint. A similar trend was also observed in the FEM results. Further, the wall strength of the WL2 panel was observed to be smaller than that of the panels with an opening far away from the restraint, whereas the WL1 panel possessed the highest ultimate strength among the three opening locations. This complex behavior could be attributed to the effects of the wall shape (Hw/Lw) and/or opening shape (Ho/Lo).

Opening Shape

Dong-JunLee, [36] whose experimental program compared load versus maximum lateral deflection responses of wall panels with door type openings DC and DL (see figure 19 (d) for symmetric and figure 30 (e) for asymmetric door type openings) are given for one- and two-way action. These types are compared to the responses of the same size walls with window type openingW1C1.2 and W1L1.2 (figure 30 (b) for asymmetric window type openings). The ultimate failure loads of wall panels with door type opening (DC and DL) were found to be reduced compared to those with window type openings (W1C1.2 and W1L1.2) in both one- and two-way action. A decrease in axial strength ratio of walls with door type opening located at the center and left side is found 31% and 41% for one-way walls and both 26% for two-way walls respectively compared to the ratios of normal strength concrete walls with one opening located at the center (O50W1C1.2 and T50W1C1.2). Varying door type opening and location indicates an overall decrease in ultimate strength for walls.

However, the ultimate failure loads of wall panels with door type opening did not decrease much compared to the decreased size of the opening area in both one- and two-way actions; the opening area of door type (Ho.Lo = 750mm.300mm) was2.5 times larger than that of window type (Ho.Lo = 300mm.300mm).

Axial Capacity

Mehdi Mohamamdpour Lima, et al., [11], conducted 18 one-third scaled RC walls with an opening were strengthened using seven distinct externally bonded (EB) CFRP sheet layouts and were loaded up to failure under various support conditions (one-way action and two-way action with three or all four sides restrained) as presented in figure20. However, wall and opening geometries were kept constant which have square geometry with the length and height being 1200 mm and the thickness equal to 40 mm(1200 mm long, 1200 mm tall and 40 mm thick and tested up to failure, they have symmetric openings (450 mm x 450 mm). Each wall configuration exhibited a slenderness ratio (Hw/tw) of 30. The specimens were loaded by axial loads and reinforced by centrally positioned single layer of welded wire fabric reinforcement, comprising of deformed 4 mm diameter bars with 100 mm spacing in the both directions. There are hinged connections at the top and bottom boundaries of the specimen and free side edges. Steel mean yield strength (fy) was 500 MPa. CFRP sheets in OW-AF are externally bonded with thickness (0.128 mm) and tensile modulus234000 MPa. Three hydraulic jacks, each with a most extreme limit of 800 kN, were connected together to apply a uniformly distributed load, with controlled total force, along the wall length. The specimens were subjected to an eccentricity of one sixth of the wall thickness to obtain the curvature and tension side in a specific side. A steel rod was welded to both of loading beam to apply eccentric distributed loading, designed to fit into a guide system connected to the upper edge and lower edge of the specimen.

Mehdi Mohamamd pour Lima et al., [11],drew a number of conclusions from results illustrated in figure 28, some of which are outlined as follows.

- Externally bonded CFRP sheet can significantly increase the strength of RC walls.
- Varied success was achieved in ultimate strength gains under different support conditions and CFRP sheet layouts. For RC walls with OW an enhancement of ultimate strength between 14.0-59.7% was observed, while this range for walls with TW3S and TW4S were 3.0-40.8% and 18.4-24.9%, respectively.
- Considering various support conditions, the ultimate strength of RC walls with OW were 60% and 40% of that with TW3S and TW4S, respectively.

The efficiency of CFRP sheet layouts were also investigated where WF layout presented the least efficient pattern of strengthening, whereas, CF and DF were the most effectual layouts for walls in one-way and two-way action, respectively. The analysis results show that the ultimate load carrying capacity, crack patterns, principal plastic strain of concrete, and equivalent stress are considerably affected by the variation of CFRP sheet thickness. When the CFRP sheet thickness increases to 0.348 mm, the ultimate load capacity increases. Cosmin Popescu et al., [29] conducted an experimental investigation on FRP strengthened RC walls with openings under four sides restrained (TW4S). However, no design chart or formula was proposed based on their experimental outcomes. The FRP was fully wrapped around the opening with a mechanical anchorage also used as illustrated in figure 36. The test matrix can be divided into three stages, designated I-III, in which reference (unstrengthened) specimens, pre-cracked specimens strengthened by FRP and uncracked specimens strengthened by FRP (duplicated to increase the reliability of the data) were tested, respectively. As mentioned above, all of these specimens were 1800mm long, 1350 mm wide and 60 mm thick. Three specimens were loaded to failure in stage I: a solid panel, a panel with a "small" symmetric half-scaled single door-type opening (450 mm x 1050 mm), and a panel with a "large" symmetric half-scaled double door-type opening (900 mm x 1050 mm). The small and large openings represent 25%and 50% reductions, respectively, in the cross-sectional area of the solid wall. Thus, these tests enabled evaluation of effects of introducing new openings in a solid wall.

The damage level was evaluated in terms of ultimate load, crack pattern, displacement profiles, and strains in concrete and steel reinforcement, ductility, and energy release at failure. In stage II, two specimens (one with a small opening and one with a large opening). To create cracks of this width the specimens were loaded up to 75% of their

unstrengthened axial capacity. They were subsequently completely unloaded then strengthened by FRP and tested to failure. This procedure mimics scenarios in which the creation of openings and subsequent presence of a sustained load results in degradation of a wall. In stage III duplicated specimens with openings of each size were strengthened with the FRP system in an uncracked state then loaded to failure. For convenience, the specimens are designated according to the stage when they were tested(I, II or III), their type (C, S or L: for solid wall, and walls with small and large openings, respectively) and (for specimens used in stage III) serial number. As shown in figure 37, FRP-confinement and mechanical anchorages increased the axial capacity of walls with small and large openings (which had 25% and 50%reductions in cross-sectional area, respectively) by 34-50% and 13-27%. This enhancement in ultimate failure load was up to 85-94.8% and 56.5- 63.4% of the corresponding solid wall. Similarly, concrete crushing accompanied by de-bonding of the FRP sheet occurred at failure.

Crack Pattern

Bashar Mohammed et al., [23] whose experimental program was mentioned in section 2.2.1, revealed that, wall panels with openings strengthened with CFRP displayed different crack pattern compared to the wall panels without CFRP. Different patterns of CFRP sheet strengthening. Method show different crack patterns as well. Figure 38 shows some crack patterns for the wall panels with openings strengthened with CFRP sheet pattern 1 (90°), where the CFRP sheet applied along the wall panel opening. The failures occurred in tension face whereby the CFRP will either be ruptured or torn from the concrete when the force applied is greater than the CFRP tension capacity. The failure of concrete took place after the CFRP have been torn from the concrete surface. The advantages of applying CFRP sheet along the wall panel opening is that the wall panels will only fail in column strips, either near the center of the wall or horizontally from the opening corner. Figure 24 shows some crack patterns for wall panels with openings strengthened with CFRP sheet pattern 2. The crack patterns for wall panels in this batch are similar to wall panels with openings without CFRP. The cracks were initiated vertically from the top or bottom of the wall panel opening towards upper or lower support, followed by horizontal cracks at the column strips. The cracks at the column strips were not originated from the opening corners. The cracks that happened near the applied CFRP sheet will go around the CFRP because the CFRP resisting the force. Figures 26 shows maximum principal plastic strain of concrete. Hussam K. Risan,et al., [41], whose F.E. program stated that it is obvious from these figures that general cracking pattern and plastic strain tendencies of concrete were similar for one way and two way actions of the RC wall for CFRP thicknesses ranging from 0.128 mm to 0.512 mm, with better crack distribution with an increase in CFRP sheet thickness.

SUMMARY AND CONCLUSIONS

The review focused on the previous experimental programs. The previous researches studied the impact of opening on the R.C. wall and there are very few researches which study the opening size, orientation, shape ,aspect ratio and optimum method to strength in R.C. wall to provide the best performance and that is found as a problem. It is evidence that the most effective strengthening scheme to increase the axial load capacity is using with one wide externally bonded CFRP sheet instead of re-arranging it to equal spaced strips which have the same total sheet area, increasing the CFRP sheet development length increases the axial capacity until limit. After exceeding that limit, the increased CFRP sheet development length will not be feasible, the increase of the CFRP sheet width leads to decrease of tensile stress concentrations value. It is also recommended to increase CFRP development length paralleled to the load direction to about 22% of the opening height. The results demonstrated that opening shape, orientation, aspect ratio and area should be taken into consideration as they affect the axial capacity of the wall ,by the compared of horizontal and vertical rectangular opening shape, the R.C wall with vertical rectangular has the highest axial load capacity which is horizontal direction decreases the wall ductility and the axial capacity values as R.C wall with vertical rectangular opening has better performance than horizontal rectangular opening, increasing the aspect ratio of horizontal rectangular openings in R.C. walls with constant opening size leads to more axial capacity reduction. Based on F.E.M and verifying it with experimental programs, by using finite element software ANSYS model found the similarity between both F.E.M. and experimental results in the term of load capacity, crack pattern and lateral

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displacement. Finite Element Method becomes an essential approach in analyzing structural engineering problems numerically. Now we can make various models with different parameters in short time by using ANSYS instead of doing it experimentally, which consumes a lot of time and money. This review may be useful for improving existing design models and to be applied in practice, enable the designer to design structures that will behave better during an earthquake and saved from collapse. The output result can be used in many countries

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Figure 1: Hysteresis curve and observed damage at the end of test - specimen N1-,[76]	Figure 2: Hysteresis curve and observed damage at the end of test - specimen M1-[76]
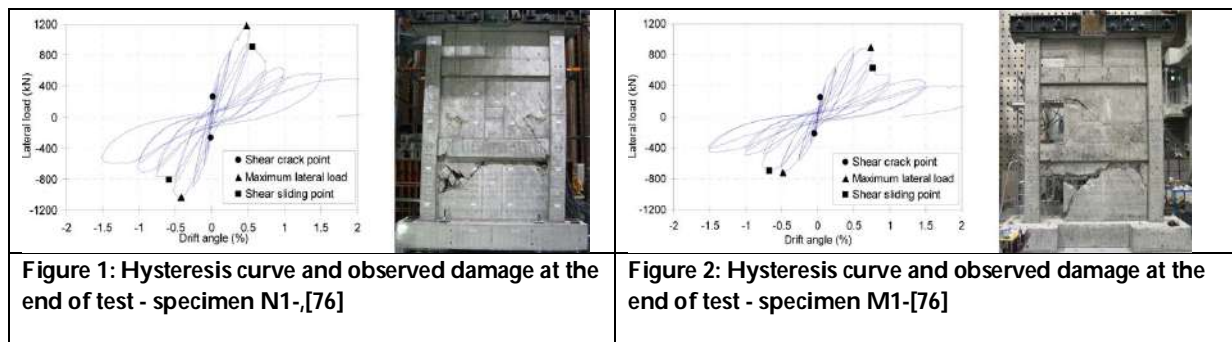
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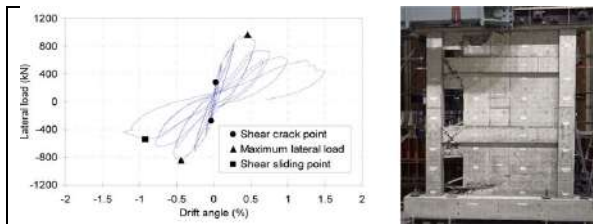
Figure 3: Hysteresis curve and observed damage at the end of test - specimen S1-,[76]
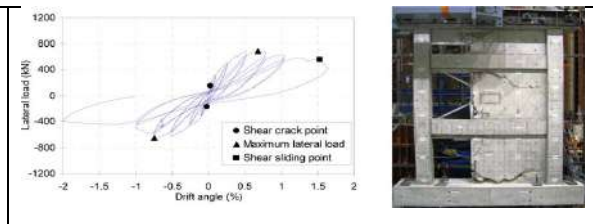
Figure 4: Hysteresis curve and observed damage at the end of test - specimen L1-[76]

Figure 5: Test Setup of unstrenghthend & strenghthend specimens [23]
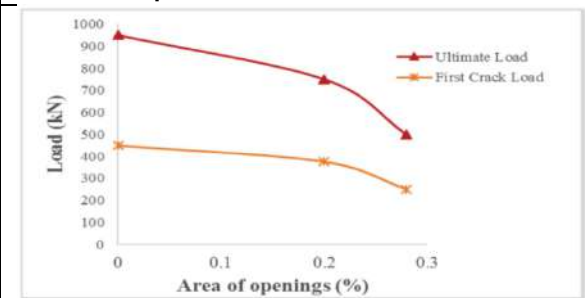
Figure 6: Variation of first crack load and ultimate load for wall panels specimens [31]
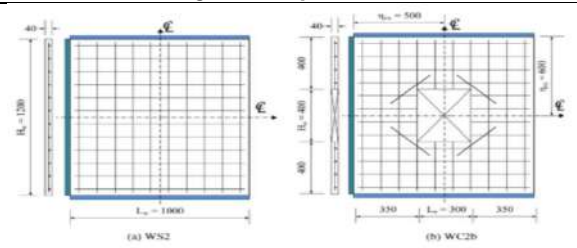
Figure 7: Typical wall panels dimensions (dimensions in millimeter) [42]
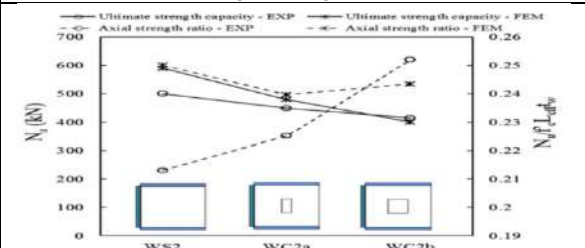
Figure 8: Ultimate loads (left vertical axis) and axial strength ratios (right vertical axis) of panels with Hw/tw = 30, Hw/Lw = 1.20. FEM = Finite Element Method [42]
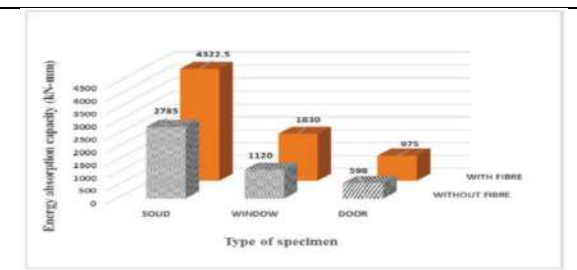
Figure 9: Energy absorption capacities of all specimens [31]
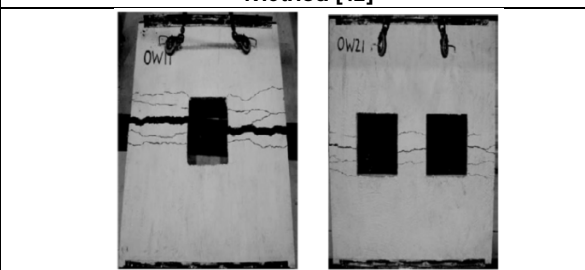
Figure 10: Crack pattern in one-way walls with openings [16]

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Figure 11: Crack pattern in two-way walls with one opening [16]

Figure 12: Crack pattern in two-way walls with two openings [16]
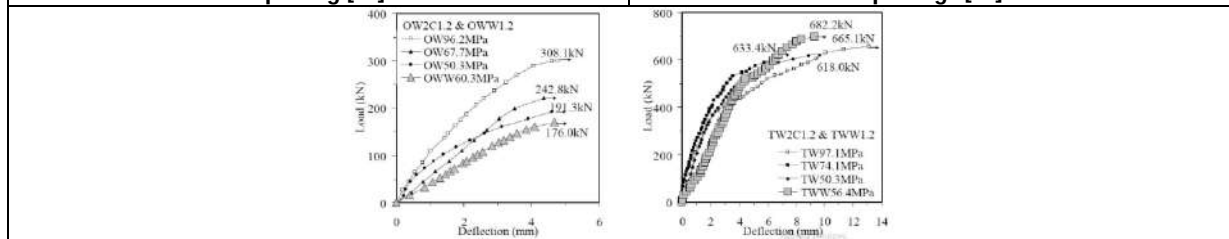
Figure 13: Comparisons of load behaviours of walls with two openings and wide opening [36]
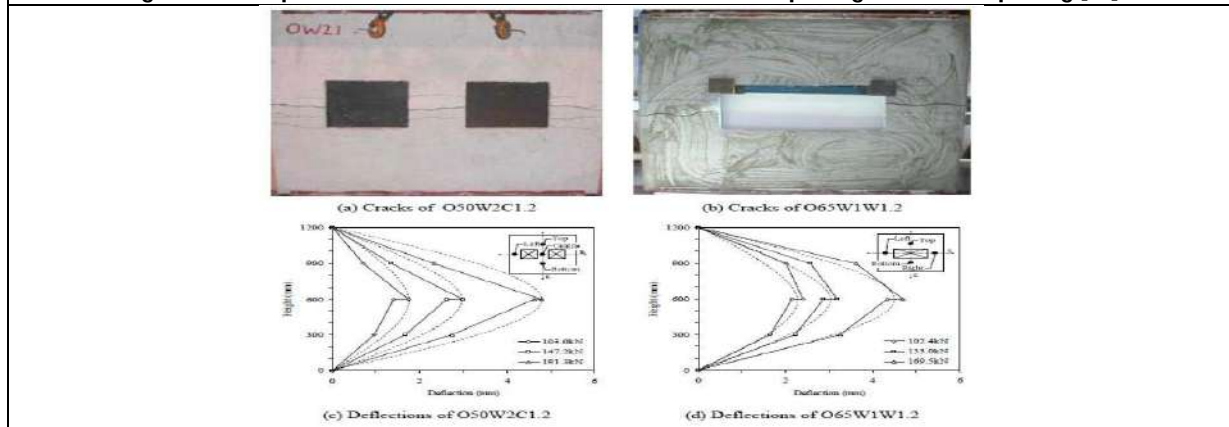
Figure 14: Behaviours of walls with two openings and wide opening in one way action [36]
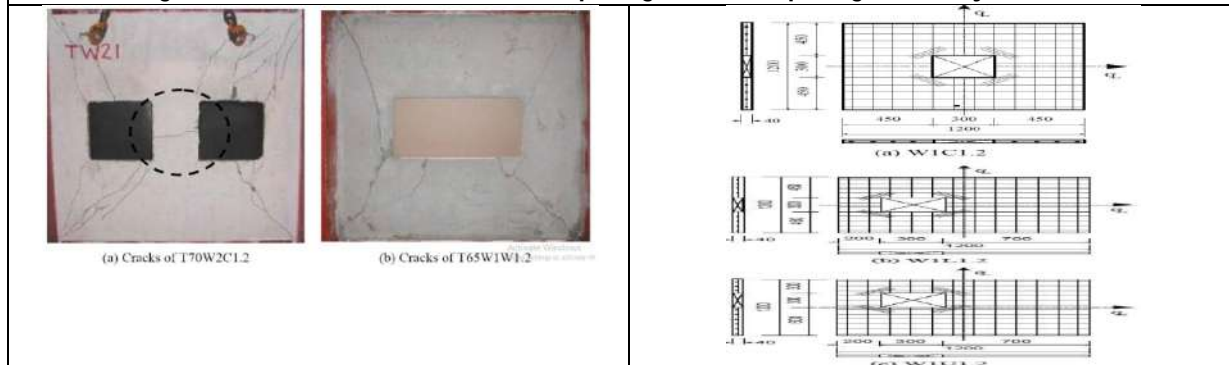
Figure 15: Behaviours of walls with two openings and wide opening in two-way action[36]

Figure 16: Reinforcement and opening layouts of test panels (units=mm)[36]

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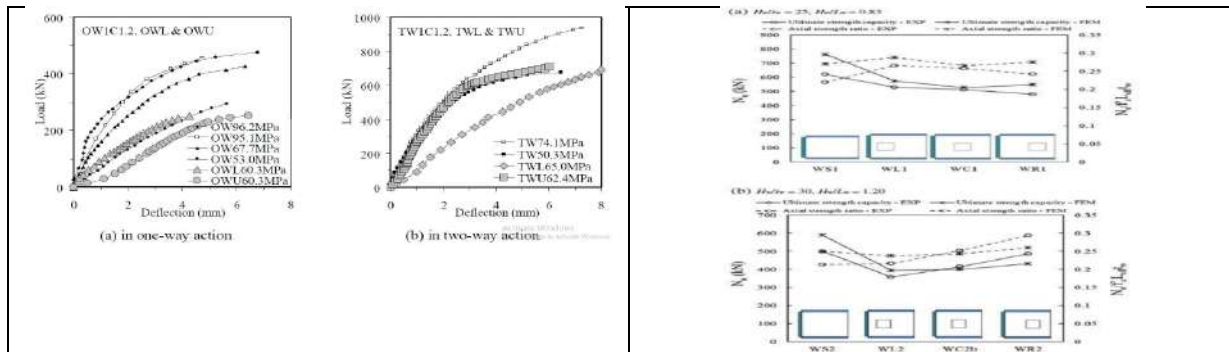
Figure 17: Comparisons of responses of walls with symmetric and asymmetric openings [36]

Figure 18: Ultimate loads and axial strength ratios of panels. FEM = Finite Element Method [42]
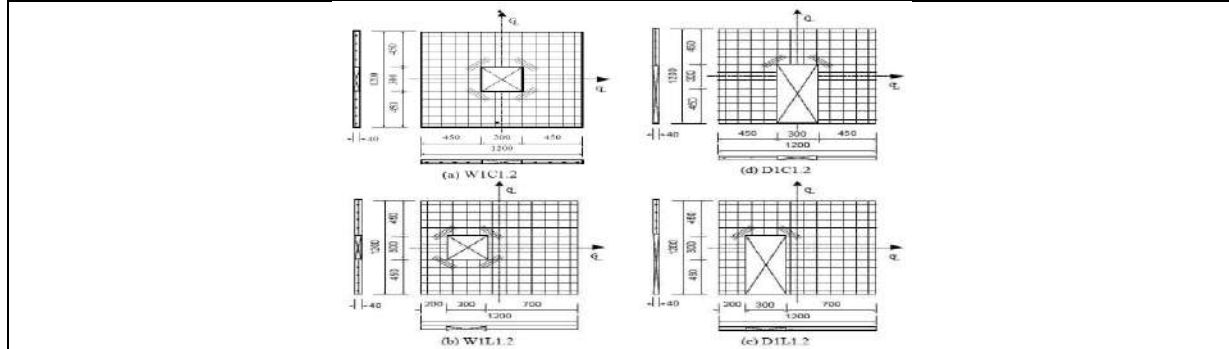
Figure 19: Opening layouts of test panels (units=mm) [36]
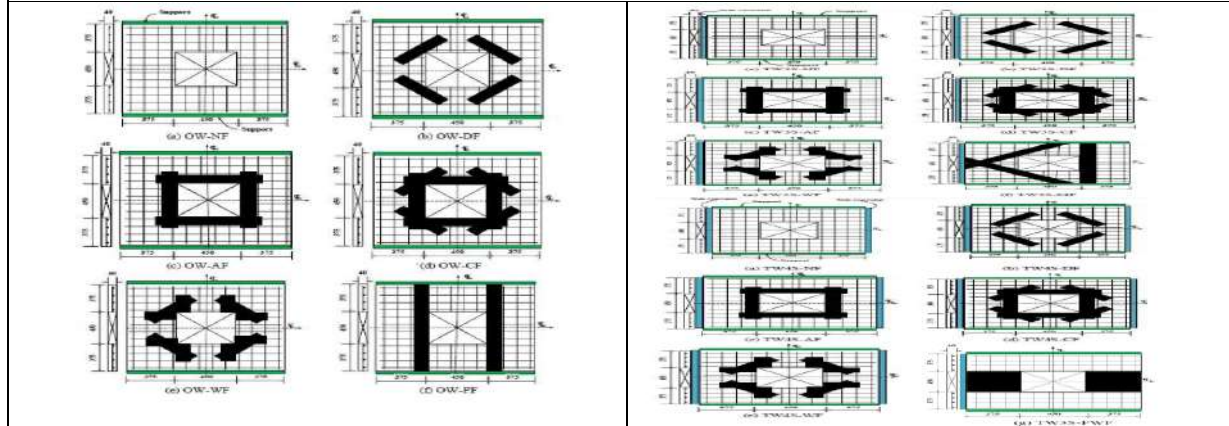
Figure 20: Panel designation and CFRP layout for walls with OW, TW3S & TW4S (dimensions in mm) [11]

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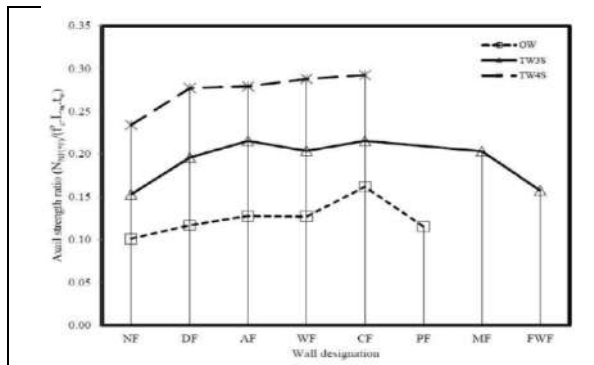
Figure 21: Axial strength ratio versus CFRP layouts [11]
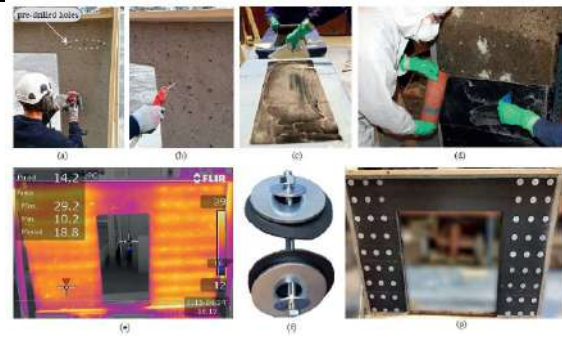
Figure 22: Strengthening process: (a) grinding the concrete surface, (b)cleaning with compressed air, (c) impregnating the fibers, (d) applying the fibersto the specimen, (e) thermal image indicating positions of the holes, (f)mechanical anchorage, (g) specimen prepared for testing [29]
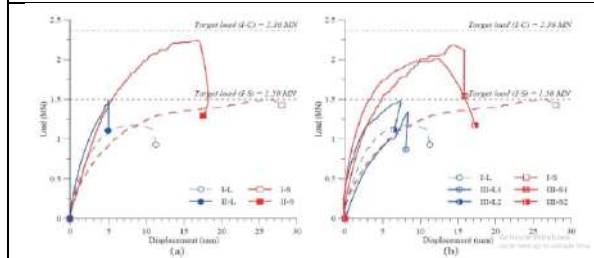
Figure 23: Load-dis. curves for ref. (stage I) specimens & (a) pre-cracked strengthened (stage II) specimens & b) uncracked strengthened specimens (stage III) [29]

Figure 24: Crack patterns on the tension face after failure of the 40mm thick wall panels with openings strengthened with CFRP pattern 1. [23]
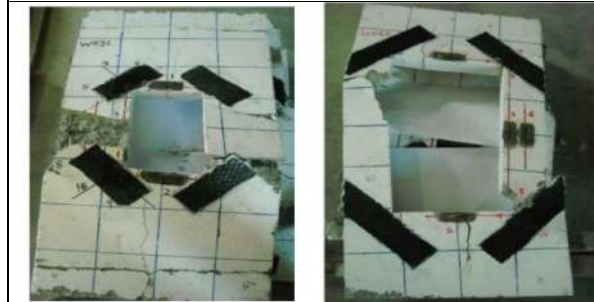
Figure 25: Crack patterns on the tension face after failure of the 40mm thickwall panels with openings strengthened with CFRP pattern 2. [23]
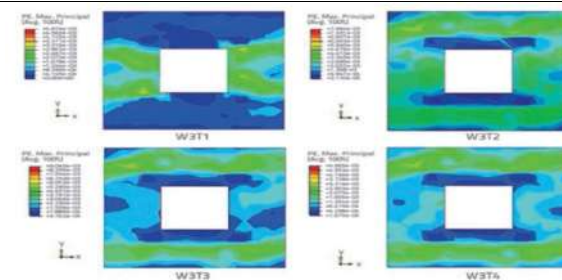
Figure 26: Maximum plastic strain for one-way action wall with around and different CFRP sheet thicknesses [41]

RESEARCH ARTICLE

Salivary Metabolic Hormones as Biomarkers of Childhood Obesity
Hormones in Childhood Obesity

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ABSTRACT

Obesity is a major threat to the health and development of children. While hormones in serum samples have been extensively investigated in childhood obesity research, our study attempts to explore salivary metabolic hormones as biomarkers of childhood obesity.Obese (n=83) and non-obese (n=83) school children aged 6-11 years were recruited as cases and controls, respectively, for this case-control study. Salivary concentrations of the hormones, Ghrelin, Leptin, Adiponectin and Insulin were assessed and compared between obese and non-obese children to evaluate their role in childhood obesity. The mean ghrelin (1.69 ± 0.38 pg/ml) and adiponectin (95.53 ± 64.92 ng/ml) levels of the non-obese children were higher than the mean values of the obese, while the mean values of the hormones leptin (265.95 ± 83.16 pg/ml) and insulin (224.56 ± 184.86 pg/ml) were found to be higher among the obese. Higher levels of the hormones insulin (OR: 1.09; 95% CI: 1.05-1.12) and leptin (OR: 1.03; 95% CI: 1.02-1.04) posed higher risks for childhood obesity, followed by adiponectin, which was found to be lower among the cases (OR: 0.99; 95% CI: 0.99995-0.99998) which increased the risk of childhood obesity.This study demonstrates the efficacy of saliva as a promising non-invasive tool in childhood obesity research and highlights the significance of appetite regulatory hormones as biomarkers of childhood obesity.

Keywords: Saliva, Appetite, Metabolism, Hormones, Childhood Obesity

INTRODUCTION

Childhood obesity is a public health burden worldwide. It is alarming to see the increasing rates of childhood obesity in developing countries like India. Childhood subsists to be a sensitive period for neurological, endocrine, and metabolic development. Obesity evolving at a young age contributes to an increased risk of diabetes, hypertension, and cardiovascular disease in adulthood [1].Obesity is a multifaceted problem with many contributing factors including genetics, hormone levels, overconsumption of food and sedentary lifestyle. Appetite regulation is a complex process involving communication between the hypothalamus within the brain, various gastrointestinal organs (including the stomach, the pancreas, and the intestines) and adipose tissue. Satiation (the signal that causes one to stop eating) may be initiated by neural input from the stomach to the brain, signaling gastric distension after food consumption. This is quickly followed by the release of various hormones sensing the digestion and absorption of nutrients and initiate satiety (the feeling of fullness that persists after eating). The gastrointestinal tract is the largest endocrine organ in the body and plays an important appetite-regulating role as a source of numerous regulatory peptide hormones. When the levels of the hormones inducing hunger increase, the demand for food intake increases, thereby contributing to excess energy storage and increased levels of fat, all of which sum up to the cause of obesity [2].

While most of the metabolic biomarker analysis in the field of obesity is performed using blood samples, studies on salivary biomarker analysis seem to be scarce. The salivary proteome consists of thousands of proteins, including hormonal mediators of energy balance. Studies state that human salivary glands can produce and release ghrelin and have also observed a significant correlation with serum or plasma ghrelin levels [3]. Saliva could be a boon in pediatric populations considering the parental apprehension towards withdrawal of blood samples from children. Salivary biomarkers have also been reported to be an effective and sustainable alternative due to the ease of tissue access, cost-efficiency and the ability to collect it in multiple settings, repetitively [4]. In comparison to collection of blood, saliva is associated with lower infection rates, decreased cost, increased patient acceptance, and higher participant compliance. Saliva also has the advantage of offering insight into the gastrointestinal tract, which could be useful when examining obesity [1]. Accordingly, in our study, we have adopted saliva as the medium of biomarker analysis among children. The biomarkers of interest were appetite regulatory hormones such as ghrelin, leptin, adiponectin and insulin. Although there are several studies in both children and adults on the causes of obesity, little is known about the physiological role of appetite regulatory hormones in humans, especially children. Hence, there is ample scope for research in this area [2]. In this pretext, the framework for our study was developed to evaluate the significance of salivary metabolic hormones as biomarkers of childhood obesity.

MATERIALS AND METHODS

Study Design
A case-control study design was adopted in this study. Obese and non-obese school children of the age group (6-11 years) were the subjects of interest in this study. The levels of salivary metabolic hormones – Ghrelin, Leptin, Adiponectin and Insulin in obese and non-obese subjects were assessed and compared to determine the significance of these hormones as biomarkers of childhood obesity.

Ethical Approval
All procedures performed in this study were in accordance with the ethical standards of the institutional and national research committee (Indian Council of Medical Research) and with the 1964 Helsinki Declaration and its later amendments or comparable ethical standards. The study was approved by the Institutional Ethics Committee of Sri Ramachandra Institute of Higher Education and Research (IEC-NI/15/FEB/45/07). Written informed consent was obtained from the parents of the children who were willing to allow their children participate in the study.

Setting

The study was conducted in an elementary school and a summer campsite in Chennai, India, from July 2017 to March 2018, as part of an extensive research in the area of salivary appetite regulatory hormones and an intervention program targeting these hormones in obese children. Permission was obtained from the concerned authorities of the elementary school and the summer campsite for conducting the study. Anthropometric assessment of all children aged between six and eleven years was carried out at the sample sites to categorize them into obese and non-obese groups. Children who gave written informed parental consent and assent to participate in the study alone were recruited for the study as per the sample size recruitment.

Participants

A total of 1432 children from an elementary school (n=1378) and a summer campsite (n=54) were assessed for childhood obesity, using standard protocols. Of the 1432, 166 children (83 obese and 83 non-obese) were recruited for this study based on inclusion and exclusion criteria and consent to participate in the study. The criteria for inclusion were – i) children aged between six and eleven years categorized as normal (control) and obese (case) using the WHO growth reference for school aged children (2007) and, ii)children who gave written informed parental consent and assent to participate in the study. The criteria for exclusion were – i) children who had an obvious underlying medical cause of obesity (evaluated using a questionnaire) and, ii) children who had undergone any medical or nutritional therapy for obesity in the past six months.

Tools Used –

Anthropometry

Anthropometric measurements such as height and body weight of the subjects were assessed using standard protocols. A calibrated stadiometer was mounted on a wall and the height (in centimeter (cm)) of each subject was measured in an upright standing position on an even surface adjacent to the wall. Body weight of each subject was measured using a body fat analyzer (TANITA UM-076) and recorded in kilogram (kg). The readings were entered in the nutritional survey module of WHO AnthroPlus v.1.0.4 (2009) software. The height-for-age 'Z' (HAZ) scores, weight-for-age 'Z' (WAZ) scores, and body mass index-for-age 'Z' (BAZ) scores of the subjects were calculated using the WHO AnthroPlus software and the subjects were categorized into obese and non-obese categories based on their BAZ scores.

Salivary Analysis

Collection of samples of Saliva

On the day of sample collection, the subjects were asked to come to the sample site in a state of fasting. The subjects were made to rinse their mouth with water before sample collection. Saliva was collected using sublingual cotton roll technique (cotton roll placed under the tongue of the subject for a minute). Using a sterile forceps, the cotton roll was transferred to a 50 mL syringe and injected into a vacutainer tube, to collect approximately two milliliter of saliva from each subject. The samples collected in vacutainer tubes were arranged in a 96-vial storage rack placed in a freezer box. The principle investigator shifted the samples stored in the freezer box to the testing laboratory under safe conditions within 24 hours of sample collection. The samples were stored at -20°C and analyzed within a period of six months.

Multiplex Analysis of Salivary Markers

Magnetic Luminex® Assays were used to assess the concentration of the selected hormones in each sample. The assays (166 saliva samples) for three biomarkers leptin, adiponectin and insulin were performed on 50 µl of saliva sample using premixed 3-plex magnetic bead panels on a Bioplex 200 platform with no dilution. The assay procedure was carried out following the manufacturers' protocol. The results were read within 90 minutes post assay and evaluated using an analyzer.

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Ghrelin Assay

As inclusion of ghrelin in the premixed kit used for the other markers was not possible due to the interference in magnetic bead regions, a separate test kit (RayBio® Ghrelin Enzyme Immunoassay (EIA) Kit: EIA-GHR, EIAM-GHR, EIAR-GHR) was used to assess ghrelin in 100 µl of each saliva sample. The assay employed an in vitro quantitative technique for detecting ghrelin peptides based on competitive enzyme immunoassay principle. The assay procedure was carried out following the manufacturers' protocol. A standard curve of known concentration of Ghrelin peptide in the samples was assessed.

Sample size

166 (Obese, Cases = 83; Non-obese, Controls = 83) was the calculated sample size, assuming $\mu1=48.0$ and $\mu2 = 63.1$; Difference of means = 24.6, based on the study conducted by Gil *et al.*, 2009 [5], at a Power of 80%. The sample size was calculated using Piface by Russell V. Lenth. Version 1.76 – 29 June 2011.

Statistical analysis

All descriptive statistics are expressed using mean and standard deviation after analyzing the data for normality, confirming the normal distribution of data. Independent sample t-test was used to test the mean difference between the control and case groups. Box and whisker plots were drawn to display the variation in hormone levels of the subjects through the quartiles of data. Univariate binary logistic regression was used to calculate the risk factor analysis. Odds ratio was calculated to evaluate the odds of salivary metabolic hormones as biomarkers among the obese and non-obese groups and to imply an association between the salivary hormones and childhood obesity. The statistical analyses were done using SPSS version 23.0 and any p - value less than 0.05 was considered as statistically significant.

RESULTS

Of the 1432 children assessed for enrollment in the study, 758 (256 obese and 502 non-obese) fulfilled the inclusion criteria, out of which 592 subjects were excluded from the study due to dissent to participate and randomized selection. Finally, a total of 166 subjects (83 obese and 83 non-obese) participated in the study and there were no drop outs, as the study involved a one-time diagnostic observation. Study report flowchart according to the STROBE (Strengthening the Reporting of Observational Studies in Epidemiology) is presented in Figure 1. The mean values of baseline characteristics of the subjects are mentioned in table 1. The mean age of the subjects was 8.65 ± 1.06 years. With respect to anthropometry, the mean HAZ of the subjects in control group and case group was found to be 135.07 ± 7.14 and 133.76 ± 6.23, respectively. The mean WAZ of the control group was found to be 0.07 ± 0.94 and that of the case group was 1.72±0.39 (p value <0.001). The mean BAZ of the subjects belonging to the control group was found to be –0.13±0.93 and that of the cases was found to be 2.11 ± 0.28 (p value <0.001). .

Figure 2 pictorially represents the mean salivary concentrations of metabolic hormones of the subjects using box and whisker plots. Mean salivary ghrelin values of obese and non-obese children were, 0.77±0.28 pg/ml and 1.69±0.38 pg/ml, respectively. Mean salivary adiponectin values of obese and non-obese groups were, 28.74±22.06 ng/ml and 95.63±65.92 ng/ml, respectively. The mean concentrations of the hormones leptin among obese and non-obese groups were, 265.95±83.16 pg/ml and 136.99±72.48 pg/ml, respectively. Mean salivary insulin concentrations of obese and non-obese children were, 224.56±184.86 pg/ml and 47.64±23.76 pg/ml, respectively. The statistical evaluation of mean difference of salivary metabolic hormones between obese and non-obese groups revealed that there was a significant difference in the concentration of salivary biomarkers (P <0.001). The odds ratio is a measure of relative risk and is usually calculated by dividing the odds of exposure among the cases by the odds of exposure among the controls [6]. In this study, the exposure was considered to be salivary metabolic hormone concentrations, in order to investigate the odds of these hormones as risk factors for childhood obesity. The results of the univariate binary logistic regression analysis highlighted the significance of salivary metabolic hormones as biomarkers of childhood obesity. It reveals that, higher levels of the hormones insulin (OR: 1.09; 95% CI: 1.05-1.12) and leptin (OR: 1.03; 95% CI: 1.02-

1.04) pose higher risks for childhood obesity, followed by adiponectin, which was found to be lower among the cases (OR:0.99; 95%CI:0.99995-0.99998) increasing the risk for childhood obesity (Table 2).

DISCUSSION

The gut–brain axis harbors a pivotal role in the regulation of food intake and the maintenance of body weight. A complex array of signals from peripheral and central nervous systems, likely under epigenetic programming influences psychological and social factors to determine energy balance and body weight homeostasis [7]. The cluster of hormones that regulate appetite and food intake is wide-ranging. When the levels of the hormones inducing hunger increase, the demand for food intake increases, thereby contributing to excess energy storage and increased levels of fat, all of which sum up to the cause of obesity. Belfort-DeAguiar and Seo (2018) concluded that, obese individuals have elevated insulin and leptin levels and decreased ghrelin levels in comparison to normal weight individuals [8]. They have affirmed that insulin and leptin levels parallel body weight status, and insulin and leptin resistance play an indicative role in the pathogenesis of obesity. Thanakun *et al.,* (2014) and Li *et al.,* (2010) found significant correlation between salivary and serum ghrelin and adiponectin levels, suggesting salivary ghrelin could be a possible alternative to serum ghrelin as a biomarker in predicting the risk of childhood obesity [9,10]. In our study we observed a significant difference in the concentration of salivary metabolic hormones among the obese and non-obese subjects. The results indicated that the mean concentrations of ghrelin and adiponectin of non-obese group were higher than the mean values of obese, while the mean values of the hormones leptin and insulin were found to be higher among obese when compared to the non-obese controls. Several studies on serum analysis of appetite hormones highlight that obese children show significant lower adiponectin and ghrelin concentrations and higher insulin and leptin levels [11-13]. The findings of the study conducted by Goodson *et al.,* (2014) on metabolic disease risk in children using salivary biomarkers were consonant with the results of ours, presenting lower levels of ghrelin and adiponectin in the obese group when compared to the normal subjects and higher levels of insulin and leptin in the obese group [14].

The mechanism behind the downregulation of ghrelin in obese children could be attributed to their elevated leptin or insulin levels, as studies show that fasting ghrelin levels negatively correlate with fasting insulin and leptin levels. This state represents an adaptation towards positive energy balance and increased weight gain in these children [15]. Decreased concentration of adiponectin among the obese children in our study is similar to the findings of many studies carried out in the serum samples of obese children [16-18]. Adiponectin may be one of the signals linking inflammation and obesity. Soliman *et al.,* (2012) suggest that in a majority of obese individuals, serum leptin concentrations are increased and leptin administration shows only very limited effects due to leptin resistance [19]. Leptin resistance is associated with insulin resistance and abdominal obesity. Increased appetite is associated with altered levels of appetite regulatory hormones and thus, these hormones are identified as potential neuroendocrine markers and mediators in childhood obesity, as insisted by Hagen *et al.,* (2015) [20].

CONCLUSION

Despite the need for robust research in childhood obesity, the practical challenges and risks in including children being vulnerable groups, as subjects, limit the scope of clinical research in this field. While difficulty in obtaining permissions from authorities and parents for using invasive techniques like blood withdrawal are on one hand, on the other, convincing children to such invasive methods pose a great challenge. Such difficulties form one of the major reasons for dropouts from the study. Hence, the conquest for reliable, sustainable and effective non-invasive tools of clinical research is of dire need. Saliva seems to be a promising non-invasive tool to widen the scope of clinical research in childhood obesity. The present study has thrown light on the possibility of considering salivary samples in the evaluation of metabolic hormones, which may further enable us to intervene and prevent or treat the onset or incidence of childhood obesity.

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Statements

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Table 1: Mean Baseline Characteristics of the Subjects

Characteristics	Groups (N=166)		p-value
	Obese (Case) (Mean ± S.D.) (n=83)	Non-obese (Control) (Mean ± S.D.) (n=83)	
Age	8.67 ± 1.09	8.61± 1.03	0.716
Height (cm)	133.76 ± 6.23	135.07 ± 7.14	0.211
Weight (Kg)	38.74 ± 5.59	29.62 ± 5.73	0.000**
WAZ	1.72 ± 0.40	0.07 ± 0.94	0.000**
HAZ	0.12 ± 0.46	0.40 ± 0.75	0.024*
BAZ	2.11 ± 0.28	– 0.13 ± 0.93	0.000**

**P<0.01; *P<0.05

Table 2: Salivary Metabolic Hormones as Biomarkers of Childhood Obesity (N=166)

Variables	Odds Ratio	p-value
Age	1.06 (0.79-1.41)	0.714
Gender - Female	1 (0.55-1.84)	1.000
Height	0.97 (0.93-1.02)	0.211
Weight	1.33 (1.22-1.45)	0.000**
WAZ	132.99 (20.86-847.86)	0.000**
HAZ	0.47 (0.28-0.80)	0.005*
Ghrelin	0.001 (0-0.01)	0.000**
Leptin	1.03 (1.02-1.04)	0.000**
Adiponectin	0.99 (0.99995-0.99998)	0.000**
Insulin	1.085 (1.054-1.117)	0.000**

**P<0.01; *P<0.05

Figure 1: STROBE Study Chart
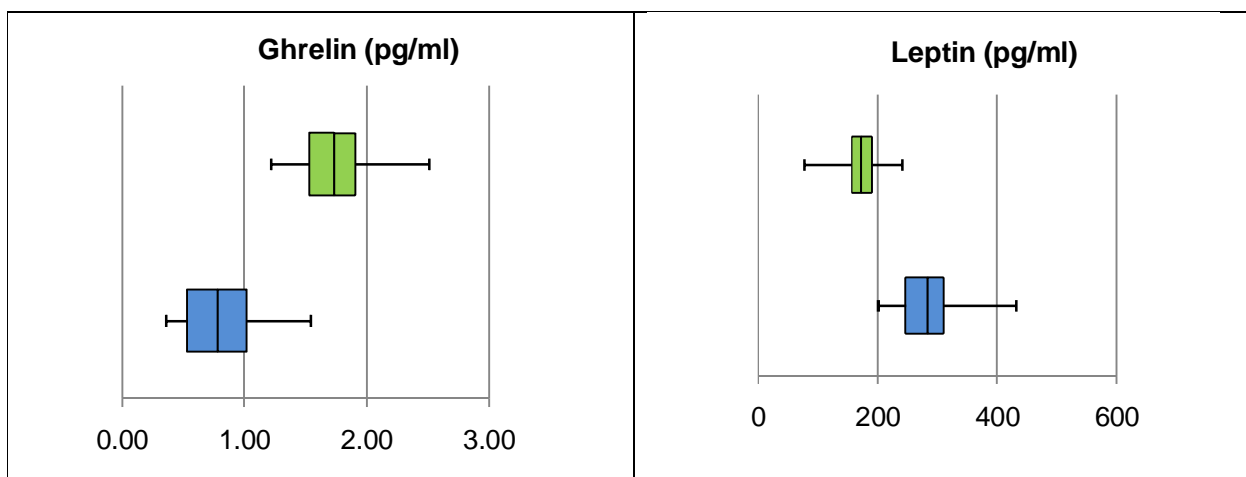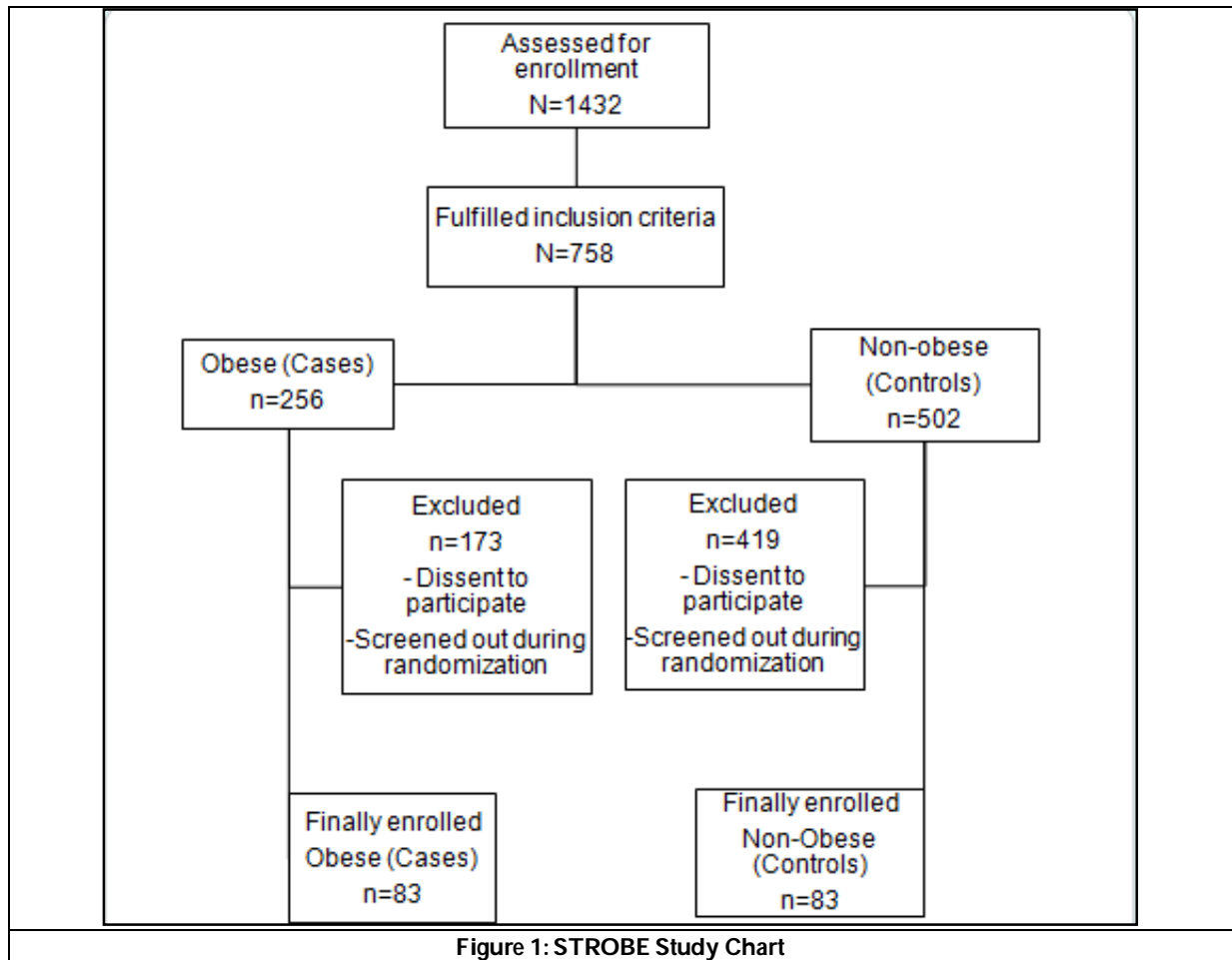
Shiny Lizia *et al.*
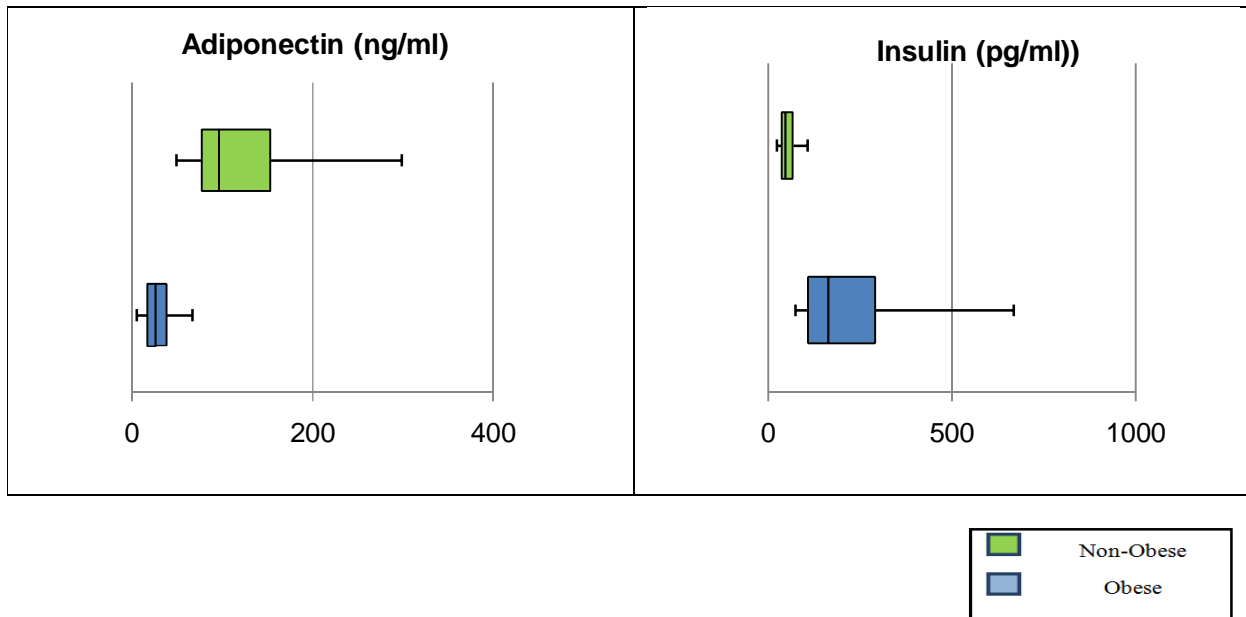
Figure 2.Mean Concentrations of Salivary Metabolic Hormones among the subjects

RESEARCH ARTICLE

Phytochemical Analysis and TLC Profiling of the Sequential Leaf Extract of *Moringa oleifera*: A Potent Medicinal Plant

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ABSTRACT

Moringa oleifera is commonly known as drumstick. It is a rich source of calcium and different types of macro and micronutrients. In the present study for phytochemicals analysis, *Moringa oleifera* leaves were used in different solvents *viz* aqueous, ethanol, ethyl acetate and benzene. The extracts shows presence of tannins, terpenoids, glycosides etc.TLC profiling of the sequential extract indicates the presence of number of phytochemicals. There was variation in the Rf values of phytochemicals in different solvent systems. Different Rf values gives an idea about their polarity. Rf values acknowledge the polarity of phytochemicals and helps in the selection of suitable solvent system. The stronger a compound is bound to the adsorbent, the slower it moves up the TLC plates. Non polar compounds move up the plate most rapidly (higher Rf value), polar substances travel up the TLC plate slowly or not at all (lower Rf value). The mixture of solvents can be separated out according to their variable polarity. This study will help in the selection of suitable solvent system for separating pure compounds by column chromatography.

Keywords: *Moringa oleifera,* phytochemicals, terpenoids, TLC.

INTRODUCTION

Moringa oleifera is a fast growing, evergreen tree with a height of 32-40 feet belongs to Brassicales and family Moringaceae of Angiosperms and commonly known as drumstick [1] Its leaves, flower seeds and soft pods are consumed as they have nutritive and medicinal properties [2]The leaves of *Moringa oleifera* have low calorific value so they are used in diet of obese [3].The phytochemical investigation of powdered leaves shows the presence of glycosides tannins alkaloids etc.[4]It is rich in macro- and micronutrients and other bioactive compounds which are important for normal functioning of the body and prevention of certain diseases.[5,6] Leaves, flowers, seeds, and almost all parts of this tree are edible and have immense therapeutic properties including antidiabetic, anticancer, antiulcer, antimicrobial and antioxidant.[7]Medicinal plants are the sources of drugs of traditional system of medicines, modern medicines, food supplements, chemical entities for synthetic drugs. [8-12]*Moringa oleifera* also considered as a medicinal plant which hows the antifungal and antimicrobial properties [13,14]. Studies also shows that different phytochemicals compounds present in the plant are used in food industry, water treatment, fuels and pharmacy and the medicinal role of these plants can be understood by analysis of biomolecules present in it. [15]Therefore, the present study deals with phytochemical investigation and thin layer chromatography profiling of sequential extract of *Moringa oleifera.*

MATERIAL AND METHODS

Collection of Samples: The experimental plants *Moringa oleifera* leaves were collected from the Government Garden and Nursery, Village-Ratgal, Kurukshetra in the month of February 2021.

Preparation of Samples: The leaves of *Moringa oleifera* were shade dried for two weeks. The dried leaves were powdered with the help of mixer grinder. Different solvents were used for preparation of extract such as ethanol, ethyl acetate and benzene.

Preparation Of Plant Extract: 50 ml of each solvent *viz*, water, ethanol, ethyl acetate and Benzene were taken and 0.5 g of the dried powder of leaves of *Moringa oleifera* were added in separate flask. The flasks were kept in shaker for 16 hours. The samples were centrifuged at 5000 rpm. The supernatant was used for further studies. The extracts obtained were stored in falcon tubes or conical tubes.

Tests For Phytochemical Constituents

Qualitative phytochemical analysis of *Moringa oleifera* was carried out to identify the secondary metabolites present in the plant extract.

Test for Glycoside: 2 ml filtered extract 1 ml of glacial acetic acid, 2 ml ferric chloride and 2 ml of conc. sulphuric acid was added. Brown color indicated the presence of glycosides.

Test for Terpenoids (Salkowski Test): 5 ml of extract was mixed with 2 ml of chloroform and concentrated sulphuric acid to form a layer. A reddish-brown coloration of the interface showed the presence of terpenoids.

Test for Tannins: 0.5 mg of dried powdered samples was boiled in 20 ml of water in test tubes then filtered. 2-3 drops of 0.1% ferric chloride were added and observed for brownish green or blue-black coloration.

Test for Alkaloids (Mayer's Test): 2 ml of the filtrates, a drop of Mayer's reagent was added by the side of the test tube. A creamy or white precipitate indicated positive test.

Thin Layer Chromatography Of Different Extract

Thin Layer chromatography was performed using three different solvent extracts, ethanol, ethyl acetate and Benzene.The thin layer chromatography works on the principle of partition or absorption chromatography. The components get separated according to its affinity with stationary or mobile phase.**Stationary phase**-Silica gel 60(G) is used as stationary phase. **Mobile phase**- Hexane: Ethyl acetate(3:1), Hexane: Acetic acid(9:1), Toluene: Ethyl acetate(4:1) **Rf**= Distance travelled by the solute/Distance travelled by the solvent front TLC plates

RESULTS

Test for the presence of different phytochemicals shows the maximum presence of glycosides in all the solvents except ethyl acetate. Terpenoids were maximum in ethanolic extract. Tannins presence was observed in aqueous extract.

Thin Layer Chromatography

TLC of all the extracts of *Moringa oleifera* was carried out in solvent phases such as Hexane: Ethyl acetate(3:1), Hexane: Acetic acid(9:1), Toluene: Ethyl acetate(4:1) TLC of ethanol extract of *Moringa oleifera* revealed the presence of 5 compounds having Rf values of 0.92,0.90,0.77,0.73 and 0.68 respectively when a solvent phase of Hexane: Ethyl acetate(3:1)was used (table 2). In solvent phase Hexane: Acetic acid(9:1), 5 spots were obtained having Rf of 0.84,0.24,0.19,0.13 and 0.07 respectively (table 3). When Toluene: Ethyl acetate(4:1) was used as mobile phase only 3 spots were obtained of Rf value 0.93,0.80 and 0.77 respectively(table 4).

TLC of ethyl acetate extract of *Moringa oleifera* revealed the presence of 5 compounds having Rf values of 0.82,0.79,0.71,0.66 and 0.58 respectively when a solvent phase of Hexane: Ethyl acetate(3:1)was used (table 2). In solvent phase Hexane: Acetic acid(9:1), only 3 spots were obtained having Rf of 0.98,0.63 and 0.25 respectively (table 3).Four spots were obtained when toluene: Ethyl acetate (4:1) is used and the compounds shows a Rf value of 0.84,0.82,0.74 and0.65 respectively(table 4). TLC of benzene extract of *Moringa oleifera* revealed the presence of 4 compounds when Hexane :ethyl acetate (3:1) used and the compounds shows Rf values of 0.84,0.79,0.73 and 0.64 respectively(table 2).In solvent phase Hexane: Acetic acid(9:1), 5 spots were obtained having Rf of 0.87,0.26,0.20,0.16 and 0.15 respectively (table 3). Toluene: Ethyl acetate(4:1)shows only 3 spots with Rf value of 1.00,0.60 and 0.53 respectively(table 4).

DISCUSSION

A variety of phytochemical constituent are present in leaves extract of *Moringa oleifera*. The active constituents such as tannins, glycosides, terpenoids and alkaloids are present in various extract were obtained and similar results were obtained by.[16] Presence of all these secondary metabolites make the plant a medicinal plant which is used in treatment of various diseases. TLC analysis was performed for optimization of a specific solvent system suitable for separation of various compounds. Thin layer chromatography of different extract using mobile phases which are mixture of different solvent and shows a no. of bioactive compounds.[17] These compounds shows different Rf values comparable to those of ethanol extract of *Moringa* leaves as reported by.[2] Ethyl acetate extract also shows presence of different compounds and shows similarity with.[18]When Benzene extract was used only three compounds were obtained using toluene: Ethyl acetate as mobile phase.[19] Methanol extract of *Moringa* shows presence of phytochemical similar to ethanolic extract. [20,21]

CONCLUSION

The chemical constituent's present in plant extract are provided by the qualitative phytochemical screening of plants.In the present study, qualitative phytochemical screening showed the presence of metabolites such as tannins, terpenoids, glycosides were present in the sequential extract of *Moringa oleifera.* Alkaloid test was found negative. The presence of phytochemicals reflects the importance of medicinal plants. It could be a potential source of antioxidant and preventing oxidative stress-related degenerative diseases. All this information will help in the selection of suitable solvent system for separating pure compounds by column chromatography.

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Table 1: Phytochemical analysis with different solventspresent in *Moringa oleifera* leaves

Name of the test	Aqueous	Ethanol	Etheyl acetate	Benzene
Glycoside	++	+	-	+
Terpenoid	+	++	-	++
Tannin	+	-	-	-
Alkaloid	+	+	S+	-

Table 2: Rf values of *Moringa oleifera* leaves extract by using solvent phase Hexane: ethyl acetate (3:1)

Extract	Distance travelled by solvent(cm)	Peak obtained(cm)	Rf values	Colors of peaks
Ethanol	7.5	6.9	0.92	Dark yellow
		6.8	0.90	Light black
		5.8	0.77	Dark green
		5.5	0.73	Light green
		5.1	0.68	Yellow
Ethyl acetate	7.3	6	0.82	Dark yellow
		5.8	0.79	Light black
		5.2	0.71	Dark green
		4.8	0.66	Light green
		4.2	0.58	Yellow
Benzene	7.5	6.3	0.84	Dark yellow
		5.9	0.79	Black green
		5.5	0.73	Light green
		4.8	0.64	Yellow

Table 3: Rf values of *Moringa oleifera*leaves extract in solvent phase Hexane: Acetic acid (9:1)

Extract	Distance travelled by solvent(cm)	Peak obtained(cm)	Rf values	Colors of peaks
Ethanol	6.7	5.6	0.84	Dark yellow
		1.6	0.24	Black
		1.3	0.19	Dark greenish black
		0.9	0.13	Yellow green
		0.5	0.07	Light green
Ethyl acetate	5.5	5.4	0.98	Dark yellow
		3.5	0.63	Green
		1.4	0.25	Dark greenish orange
Benzene	6.1	5.3	0.87	Dark yellow
		1.6	0.26	Black
		1.2	0.20	Green
		1	0.16	Light yellow
		0.9	0.15	Light green

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Table 4: Rf values of *Moringa oleifera* leaves in solvent phase Toluene: ethyl acetate (4:1)

Extract	Distance travelled by solvent(cm)	Peak obtained(cm)	Rf values	Colors of peaks
Ethanol	7.5	7	0.93	Dark green
		6.8	0.80	Dark yellow
		5	0.77	Yellowish green
Ethyl acetate	7.4	6.2	0.84	Dark yellow
		6.1	0.82	Dark green
		5.5	0.74	Green
		4.8	0.65	Yellow
Benzene	7.5	7.5	1.00	Dark yellow
		6.0	0.60	Dark green
		4.0	0.53	Yellow

RESEARCH ARTICLE

Nutritional Analysis and Antioxidant Property of *Hibiscus* Extract Incorporated Immuno-Booster Jellies

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ABSTRACT

Natural plant products extensive used nowadays due to growing the load of diseases. *Hibiscus sabdariffa* (Family Malvaceous) is a plant that's cosmopolitan at some point of the planet. Its leaves, bark, roots, and vegetation are applied with inside the historical Indian device as drug to deal with several diseases. A Fruit jelly is a product made from drinkable, sugar, gelling agents, and acids in method of cooking, aiming to get gelatine like consistency. Jellies are considered to be Sensory analysis issues the interpretation of what the senses inform regarding the merchandise. It is necessary to form an inventory of descriptors adequately denoting the products and their properties. Many trails were developed and therefore the most acceptable treatment was T2 (2.5g of hibiscus powder). The proximate analysis of the hibiscus jelly (T2) showed the protein(2.053±0.05/100g), carbohydrate (28.23±0.065g/100g), fat (0.6±0.05g/gm),fibre(5.0933±0.272g/gm), moisture (80.84±0.040g/gm). Antioxidant activity came out to be (37.12±0.03g/gm). Determination of shelf life of the product was also done using plate count method (using MacConkey plates) and the product was found to be safe for consumption for 7 days at room temperature and for 14 days while keeping at low temperature (4 C). The value of study was conjointly determined by the assistance of value (T0 = □15.45/100g) and (T2 = □18/100g). It is a homemade organic product, they are straightforward to carry, digest, flavoury and delectable. This study offers the transient regarding the nutritional analysis and antioxidant property of hibiscus extract incorporated jellies.

Keywords: Malvaceous, consistency, antioxidant, gelatin

INTRODUCTION

Recent years have witnessed enhanced research work reported on plants and plant products. In this regard, plants with traditional therapeutic usage are being screened more efficiently–– to be considered as a substitution or as a better alternative for chemical-based food preservatives. The plant Hibiscus Rosa Sabdariffa belongs to the family Malvaceous. **It is a cosmopolitan plant that can be found in the gardens of practically everyone.** The only limitation is that the climate is somewhat warmer with mild winters. In places with a much colder climate is used indoors. Its leaf is of perennial type and in good conditions, it **can reach up to 3 meters of height.** The shape of the leaves can vary depending on the species. They are arranged alternately and you can see dark green leaves but with a bright appearance. Hibiscus species flowers are complete, bisexual, i.e., with functional male (androecium) and female (gynoecium), including stamens, carpels, and ovary. Pollination is entomophilous i.e., by insects, or cleistogamy i.e., by self or allogamy i.e., by cross-pollination. Capsules ca. 2.5 cm long, glabrous, apex beaked. Hibiscus species are susceptible to various insect pests, viruses, and fungi, affecting leaves, fruits, and roots.

Hibiscus sabdariffa Linn, is an annual herbaceous shrub, cultivated for its flowers although leaves and seeds have also been used in traditional medicine. The calyces of the plant are used as a refrigerant in the form of tea, to make jellies and jams. The plant is reported to contain proteins, fats, carbohydrates, flavonoids, acids, minerals, and vitamins. The plant has been reported to have antihypertensive, hepatoprotective, antihyperlipidemic, anticancer, and antioxidant properties. (Mahadevan et al., 2009). Hibiscus has also medicinal properties and takes part as a primary ingredient in many herbal teas. The red flowers variety is preferred in medicine. Extracts showed antibacterial, anti-oxidant, nephro- and hepato-protective, renal/diuretic effect, effects on lipid metabolism (anti-cholesterol), anti-diabetic and anti-hypertensive effects among others. (Bernd B. *et al* (2014). Hibiscus contains an abundance of antioxidants and this is believed to give it some number of protective properties against some forms of cancer like stomach cancer and leukemia. The effectualness of *Hibiscus sabdariffa* L. (HS) in the treatment of risk factors associated with cardiovascular. HS decoctions and infusions of calyxes, and on occasion leaves, are used in at least 10 countries worldwide in the treatment of hypertension and hyperlipidemia with no reported adverse events or side effects. (Hopkins AL et al, 2013).

Total cholesterol, low-density lipoprotein cholesterol (LDL-C), and triglycerides was lowest in the majority of normolipidemic, hyperlipidemic, and diabetic animal models, whereas high-density lipoprotein cholesterol (HDL-C) was generally not affected by the consumption of HS extract. Jelly is a semi-solid product. It is prepared by boiling, straining a clear solution of pectin-containing fruit extract, free from pulp is obtained, after the addition of sugar and acid, a jelly is obtained to ensure that the TSS (Total soluble of solid) of jelly should be in the range of 65-68%. There are different types of jelly already available on the market, hibiscus jelly is not a part of the local market yet. This product has high nutritional value. *Hibiscus sabdariffa* contains a fair amount of vitamin C and antioxidants. Agar powder changed into used to present it jelly like texture. Agar creates a bulking impact and is used as a laxative to useful resource weight loss; it additionally stimulates gut to create bowel movement. In region of sugar date syrup changed into used to present the product moderate sweetness. it additionally enables in enhancing the dietary first-class of product because It consists of wholesome minerals. This product has excessive organic method price. Roselle includes a respectable quantity of diet C and antioxidants. This product is, straightforward to carry, straightforward to digest, they are flavoury and delectable.

MATERIALS AND METHODS

Standardization and Preparation of immune-booster jellies
The immune-booster jellies were formulated by homogenous mixing of orange juice, Date syrup, agar powder and hibiscus powder in different proportions for estimating their acceptability. The standardized recipe for control(T0) and different treatments (T1,T2 and T3) have been given in Table 2.1 and 2.2 respectively.

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BASIC PROCEDURE
The investigation was carried out in the Nutrition Lab, Department of Nutrition and Dietetics, Faculty of Allied Health Science, Shree Guru Gobind Singh Tricentenary University, Gurugram, Delhi- NCR. The study was carried out to develop hibiscus extract incorporated jelly. The Hibiscus powder was boiled and extracted. The hibiscus extract was homogenously mixed with date syrup. Agar powder and orange juice were mixed by continuously stirring the mixture, poured into silicone moulds and allowed to set. The whole procedure of the formulation of immuno-booster jellies can be briefed into following flow-chart:
Fig. 2.2.1 Formulation of Immuno-booster jellies

Hibiscus Powder
↓
Boiled in water
↓
Straining of extract
↓
Addition of date syrup
↓
Boiling
↓
Addition of agar powder
↓
Addition of orange juice
↓
Pour the mixture in gummies mould
↓
Allow it to set
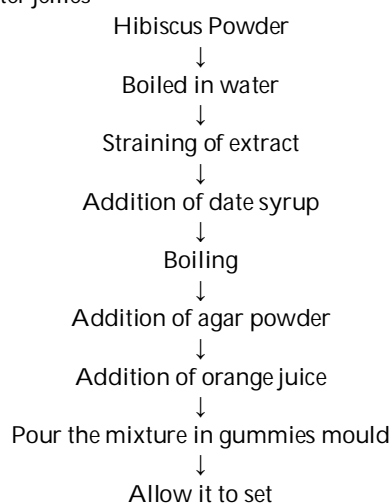
Sensory evaluation of hibiscus jelly
All the samples of developed jelly prepared with different levels of hibiscus jelly was evaluated organoleptically six times by a panel of 15 judges from the Department of Nutrition and Dietetics, SGT University, Gurugram according to the standard method of Amerine et al. (1965). The judges were served the developed jelly with one control and different test samples. The control sample (T0) was prepared from ingredients used in the standardized recipe and test samples were prepared by using hibiscus powder at different levels in the standardized recipe (T0). The samples were coded to avoid any bias. The panellists were requested to score the jelly for four sensory attributes i.e., color& appearance, texture, flavor& taste, overall acceptability by using a scorecard based on the nine-point Hedonic scale (Rangana 1986, Appendix I). The final score for each attribute for each product was obtained by averaging the score of all seven panelists.

Proximate analysis: The test was performed by Fare labs Gurugram.

 Different parameters were determined by different methods.

DETERMINATION OF SHELF-LIFE OF HIBISCUS JELLIES
The PET bottles containing hibiscus jelly after cooling were stored at both ambient(30± 2 ºC) refrigerated temperature (5 ± 2 ºC). The microbial analysis of stored hibiscus jellies were carried out at every 24 hour till 14 days.
1. Initiated with taking 2 MacConkey plates.
2. Dried the Petri plates by keeping them in a hot air oven for 10 minutes (27°C).
3. Took the plates to the working station for quadrant streaking.
4. Wire loop was sterilized using the burner for the streaking to be contamination-free.
5. Media was taken and streaking was done on the plate.
6. Both samples were streaked on the same plate. (1 plate for an incubator, 1 plate for refrigerator)
7. Place one plate in the incubator (27°C) and the other in the refrigerator (4°C) for observation.
8. Monitored the plates every 24 hours to get result accuracy

RESULT AND DISCUSSION

The analytical research activities were carried out in the laboratory Fare labs Gurugram. Two samples of each jelly were analysed in triplicate, using standard techniques and chemicals of analytical grade. The analytical values expressed on dry weight basis, were statistically analysed using t-test and ANOVA, for finding out significant differences if any. The results and discussion have been presented under the following heads:

The best product came out of the 9-point hedonic scale was Treatment 2(8.4±0.73) which had 2.5g of hibiscus powder, followed by Treatment 0(7.6±0.63) with 0g of hibiscus powder in it. Then T1 (7.3±0.9) with 1.5g amount of hibiscus powder and T3(6.13±1.88) got least score IT has 5 g of hibiscus powder in it. In a study done by Panchal *et al.* 2018, on formulation of dragon fruit jellies, the score for colour and appearance, flavour, taste, consistency, transparency and overall acceptability was highest for T3 (1000 ml fruit extract + 550 g sugar + 11 g of pectin + 0.5 % acidity) and it was 8.86 while minimum score was recorded in T6 (1000 ml fruit extract + 700 g sugar + 12.5 g of pectin + 0.5 % acidity).

Effect of hibiscus powder on sensory attributes of the developed hibiscus jellies. Sensory analysis of the developed products by the sense i.e., sight, smell, taste, touch and hearing for 5 quality attributes like colour, appearance, taste and overall acceptability, texture briefly described below:

Effect of hibiscus powder on nutrient composition of developed hibiscus jellies.
- Average protein in T2 (2.053±0.05/100g) higher than T0(1.83±0.04/100g).
- Average carbohydrates in T2 (28.23±0.065g/100g) higher than (16.15±0.0503/gm).
- Average fat in T2 (0.6±0.05g/gm) higher than T0(0.45±0.05g/100g).
- Average fibre in T2 (5.0933±0.272g/gm) higher than T0(1.273±0.05g/100g).
- Average moisture in T0 (80.84±0.040g/gm) higher than T2 (68.416±0.03/100g).
- Average antioxidant activity in T2(41.67±0.03/100g) is higher than T0(37.12±0.03g/gm).

The protein, carbohydrates, fat, fibre and antioxidant activity are high in (T2) sample due to incorporation of hibiscus powder and the moisture content is less in control sample (T0).

SHELF-LIFE OF HIBISCUS JELLIES

The sample placed in ambient temperature had microbial load of more than 103cfu/g after 7 days with no change in pH, while sample placed in refrigerator was in safe limit till day 14. Similar, results were found in case of sapota and beetroot blended jelly where the microbial count was 2 × 103 cfu/g (Gaikwad, 2016). The microbial count should not exceed to 103 per ml or g of jelly was reported by Ranganna, (2010) and Kumar and Deen, (2017) . In present findings, the microbial count had not exceeded this limit up to the day 7 in ambient temperature and day 14 in refrigerated temperature without added preservative.

CONCLUSION

This research study showed that the proximate composition of JELLIES could be enhanced through the addition of Hibiscus flower powder. The crude protein, dietary fibre, fat, carbohydrates and antioxidant activity content of the jellies significantly increased on the inclusion of hibiscus flower powder. Based on the sensory evaluation conducted, sample T2 (2.5g supplementation of hibiscus powder flower) was found to be most acceptable. The consequences showed that the product has a perfect content, protein, dietary fibre, fat, carbohydrates and antioxidant activity content, and is good for patients suffering from blood pressure issues, or people looking for good organic alternatives for hair and skin.

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Table 1. Standardized recipe (Control T0) used for development of jellies

Ingredients	Orange juice	Date syrup	Agar powder	Water
Amount	50ml	10ml	1.25g	50ml

Table 2. Treatment of hibiscus powder formulated jelly product (in 100 gm cooked preparation)

Test	Hibiscus powder	Test	Hibiscus powder
T0	0g	T2	2.5
T1	1.5g	T3	5

Table 3. Proximate analysis

PARAMETER	PROTOCOL
Protein,%bywt	IS-7219
Carbohydrates, % by wt	IS- 16 56
Fat, % by wt	AOAC 922.06
Fiber, % by wt	FLSOP/FC- 185
Moisture % by wt	IS-4333(P-2)
Antioxidant analysis:	FL/SOP/AY-39.

Table:4 Sensory characteristics of jellies with different levels of Hibiscus powder in it.

Treatment / Parameters	T0	T1	T2	T3
COLOR	8.2±1.86	8.8±1.7	9±1.2	8.9±1.2
TEXTURE	8.4±1.6	8.6±1.4	8.4±1.3	8.6±1.5
APPEARANCE	8,6±1.45	8.5±1.41	8.7±1.16	8.6±1.40
FLAVOUR	7.7±1.22	8.13±1.50	7.9±1.16	7.4±2.09
TASTE	7.4±1.30	7.8±1.56	9.2±0.7	7.2±2
OVERALL ACCEPTABILITY	8±1.27	8.06±1.09	9±0.75	7.13±1.5

Values are mean ± SD

T0	T1	T2	T3
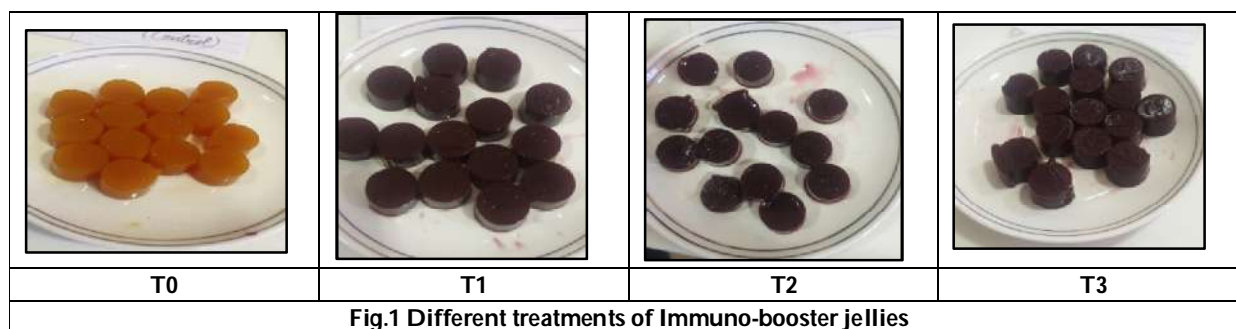
Fig.1 Different treatments of Immuno-booster jellies

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Fig. 2. (T0) Orange coloured jellySample 2. (T2) Hibiscus powder incorporated jelly
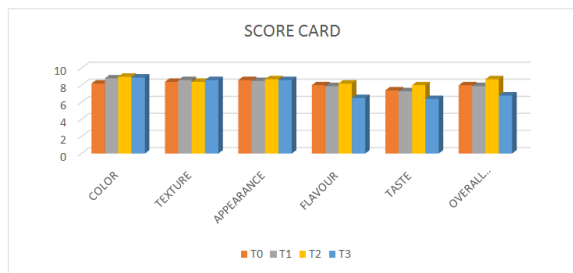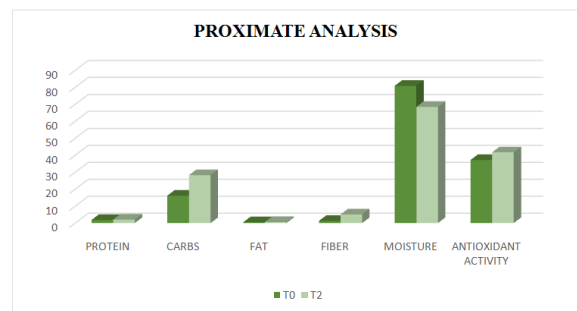
| **Fig. 3 Score Card** | **Fig. 4. Proximate Analysis** |

RESEARCH ARTICLE

Evaluation of Anticancer Efficacy of *Heliotropium curassvicum* Leaf Extract – An *In silico* Study

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ABSTRACT

Cancer is the third leading cause of death worldwide. It is a rapidly progressing disease which is characterized by uncontrolled cell division and failure of apoptosis, invasion and metastasis. Cancer in cervix is fourth leading common death cause in females. In 2006, the mortality rate of women was diagnosed as 37% by the cervical cancer in the United States. Besides in 2012, Global Cancer Observatory has reported 265,000 deaths for the annual from the cases of 527,000 and this survey portrayed the incidence of cervical cancer, leads 50% of death in the developing world. About 99.7% cases of cervical cancer are diagnosed as the infections of human papilloma virus (HPV). It has been a major risk factor of cervical cancer; commonly human genitalia affected by more than 40 genotypes of HPV been identified. Medicinal plants are being widely used in India directly in folk medicine.In spite of advances made available in allopathic medicine, so far no effective anticancer drugs are available due to its side effects. Hence for treating various diseases, alternate therapeutic use of plant need to be probably validated and documented. In the present study ethanolic leaf extract of *Heliotropium curassvicum* was used to evaluate its anti-cancer efficacy in *in silico* study.

Key words: Human Papilloma Virus, *Heliotropium curassvicum,* Cervical Cancer, Docking, *In silico* Studies

INTRODUCTION

Medicinal plants have long been used to treat and prevent a variety of diseases, infections, and infestations in domestic animals, especially livestock, around the world. About 3000 plant species are known to have medicinal properties in India. It is estimated that 40% of the world's population is directly dependent on herbal medicine for their health care [1]. In India, medicinal plants offer low cost and safe health care solutions. Several attempts were made to explore local knowledge on common medicinal plants for the treatment of diseases related to various systems of man. The sequence of secondary metabolites (SMs) and, in fact, these metabolites form the basis of many commercial pharmaceutical drugs, as well as herbal remedies derived from medicinal plants [2]**.** Worldwide, cervical cancer is the second most common malignancy in women and a major cause of morbidity and mortality. About 99.7% cases of cervical cancer are diagnosed as the infections of human papilloma virus (HPV). It has been a major risk factor of cervical cancer; commonly human genitalia affected by more than 40 genotypes of HPV been identified [3].

Chemotherapeutic drugs kill cancer cells, but also damage some normal cells, which can lead to side effects. Side effects depend on the type and amount of medication and the amount of time used for treatment. Many side effects are short-lived and decrease after treatment, but some may be long-lasting or permanent [4]. Traditional medicine is still used by about 65-80% of the world's population in developing countries as a source of early health care. Medicinal plants can be a solid source of novel treatment agents, especially for cancer. Advances in molecular biology have allowed the development of mechanism-based receptor screens to probe interactions between large molecules and find small natural product molecules as potential drug candidates in cancer chemotherapy [5]. Plant based medicines have often found important roles in the treatment of cancer and the mechanism of interaction between many phytochemicals and cancer cells are being studied extensively. The present study delineates the efficacy of the components of leaf extract of *Heliotropium curassvicum*– a lesser known medicinal plant as a potent anticervical cancer agent using *in silico* models.

MATERIALS AND METHODS

The docking analysis was performed by using AUTODOCK VINA software. Preparation of targets [6]. The crystal structure of Cervical cancer receptors p53 (PDB code = 1 OLG) and HDAC2 (PDB code = 4LY1), were retrieved from the Protein Data Bank (http://www.rcsb.org/). All bound waters, ligands and cofactors were removed from the proteins using Molegro visulaization tool and then hydrogen atoms were for optimization.

Molecular docking

Intermediary steps, such as pdbqt files for protein and ligands preparation and grid box creation were completed using Graphical User Interface program AutoDock Tools (ADT) [58].ADT assigned polar hydrogens, united atom Kollman charges, solvation parameters and fragmental volumes to the protein. AutoDock saved the prepared file in PDBQT format. AutoGrid was used for the preparation of the grid map using a grid box.

p53center_x = 0.589893701429, center_y = -1.31303979622, center_z = -0.2328
size_x = 46.2684184589, size_y = 36.3466183384, size_z = 25.04Ly1
center_x = 21.0572164097, center_y = -19.2181192038, center_z = -0.182648204102
size_x = 40.893128077, size_y = 53.274617318, size_z = 67.8313181281

A scoring grid is calculated from the ligand structure to minimize the computation time. Docking using protein and ligand information along with grid box properties in the configuration file were carried out in PyRx using Auto Dock/Vina option. Auto Dock/Vina employs iterated local search global optimizer [59,60] . During the docking procedure, both the protein and ligands are considered as rigid. The results less than 1.0 A in positional root-mean-square deviation (RMSD) was clustered together and represented by the result with the most favorable free energy of

binding. The pose with lowest energy of binding or binding affinity was extracted and aligned with receptor structure for further analysis.

RESULTS

The docking for anticancer property of 2 active compounds from *H. curassvicum* was taken. p53 and HDAC2 are the proteins used here for docking analysis. The p53 (1 olg) was docked with the 2 ligands. From the results compound 1, and 2 [Propanoicacid,3-amino-3-(4-fluorophenyl) and [1H pyrazole-4-sulfonamide,N-[(4- Fluorophenyl) methyl]-1,5-dimethyl] by forming three and two H-bonding with the amino acids in compound 1 (Gly 325, Asn 345 and Arg 333) and compound 2 (Glu 349, Arg 333). Binding energy was -6 and -5.4 kcal/mol. The HDAC2 (4LY1) was docked with the 2 ligands. From the results compound 1, and 2 [3-cyclo pentyl propionic acid] and [1H pyrazole-4-sulfonamide, N-[(4- Fluorophenyl) methyl]-1,5-dimethyl] by forming three and two H-bonding with the amino acids in compound 1 (Asp 269, Asp 181) and compound 2 (Asp 104, Tyr 308). Binding energy was -6.4 and -6.3 kcal/mol.

Control
The Hycamtin is the potent inhibitor for cervical cancer (anti-cancer) was docked against the p53 and HDAC2 target protein. Hycamtin formed 2 hydrogen bonds and interactions were made at TYR 27 at A chain and GLY 25 at A chain with the binding energy of -7 Kcal/mol.

DISCUSSION

Herbal medicine is one of the oldest forms of health care known to mankind. In Ayurveda it is also known as phytomedicine, which uses various phytoprinciples such as alkaloids, steroids, tannins and flavonoids from medicinal plants to treat various human ailments. Flavonoids, alkaloids and glycosides has shown to be responsible for cure diabetes, obesity, inflammation associated diseases, cardiovascular diseases, cancer. These phytoprinciples differ from plant to plant due to vast biodiversity. The evaluation of the anticancer activity of plant extracts is essential for safe treatment. It enables identification of the intrinsic toxicity of the plant and the effects of acute overdose [7].

Anticancer property of *Heliotropium curassvicum* was validated for cervical cancer receptors p53, HDAC2 The lead components possess similar function when compared to that of standard drug .The lead components are Propanoicacid,3-amino-3-(4-fluorophenyl), [1H pyrazole-4-sulfonamide,N-[(4- Fluorophenyl) methyl]-1,5-dimethyl, 3-cyclo pentyl propionic acid. For p53 and HDAC2 Hycamtin was used as a control drug [8]. In the present study, the docking scores against selected natural compounds showed higher binding affinity towardsp53 & HDAC 2, which is evident from the hybrid chemgauss4 score. Collectively, our results suggest that selected natural compounds from the *Heliotropium curassvicum* may inhibit the function of p53 & HADC 2 and the lead components can be considered as potential bioactive molecules to treat various p53 associated diseases [9].

CONCLUSION

Studies revealed that *Heliotropium curassvicum* has significant antineoplastic effects. The compounds present in *Heliotropium curassvicum* root extract may enhance cancer cell death via interacting with anti-apoptotic protein, p53. More extensive studies need to be done with the active compounds to elucidate the mechanism of action of the compounds against cancer cells and apoptosis.

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Table 1: Compounds from *H. curassvicum* docked with p53 (1 olg) protein

Pubchem-CID	Name of the compound	Binding energy (kacl/mol)	Interaction of amino acids	Hydrogen bonds
60699	Hycamtin	-7	TYR 27 GLY 25	2
5295512	1H pyrazole-4-sulfonamide, N-[(4 fluorophenyl)methyl]-1,5-dimethyl]	-6	GLY 325 ASN 345 ARG 333	3
579885	Propanoicacid, 3-amino -3-(4-Fluorophenyl)	-5.4	GLU 349 ARG 333	2

Table 2 : Compounds from *H. curassvicum* docked with HDAC2   (4LY1) protein

Pubchem-CID	Name of the compound	Binding energy (kacl/mol)	Interaction of amino acids	Hydrogen bonds
60699	Hycamtin	-7	TYR 27 GLY 25	2
8818	3-cyclo pentyl propionic acid	-6.4	ASP 269 ASP 181	2
5295512	1H pyrazole-4-sulfonamide, N-[(4fluorophenyl)methyl]-1,5-dimethyl]	-6.3	ASP 104 TYR 308	2

RESEARCH ARTICLE

New Classes of Odd Mean and Odd -Even Graceful Graphs for Mobile Networking Applications

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ABSTRACT

p vertices and q edges containing graph G is said to be an odd mean function when it admits $h:V \rightarrow \{0,1,2,...,2q-1\}$, f is one-one and the induced map $h^*:E \rightarrow \{1,3,5,...,2q-1\}$ defined by h* (yz) = [h(y)+h(z)]/2 when numerator is even and h* (yz) = [h(y)+h(z)+1]/2 odd function when numerator is odd. If h*(yz) satisfied the rule then it will be bi junction function. Also, the defined function odd-even graceful labeling of a graph G = (V,E) is explained as a function of f when f:V(G)→{1,3,5,…….2q+1} is an injective function. The induced function f* and f*: E (G) → {2, 4, 6 …2q} defined as f*(e) = │f (u)-f (v)│. In addition to this f*(e) is bijective for every edge e = (u, v) belongs to E (G). Hence, this paper proved that the $G_1 = P_m (QS_n)$, ∀ m≥2, n≥1, is odd mean graph, $G_2 = S_n + \overline{K_t}, n \geq 4, \forall t$ isodd-even graceful graph, The Coconut Tree $G_3 = CT(m,n)$ is odd-even graceful graph for all positive integers n and $m \geq 2$ and the jellyfish graph $G_4 = J(m,n)$ $m \geq 2, n \geq 2, m = n$ is odd-even graceful graph.

AMS Subject classification: 05C78

Keywords : Graph types, odd mean graph, odd-even graceful, star graph, Path, Quadrilateral Snake.

INTRODUCTION

Graph labeling can be applied for Mobile Adhoc Networks (MANETS) issues such as connectivity, modelling, the network and simulation, scalability and routing. Since a network can be modelled as a graph and it can be used to overcome these issues which can be represented as algebraic matrices. Also networks can be automated by means of algorithms. Systematic studies on various applications of graph labeling have been overviewed for this investigation [1-3]. The reported α – labeling (or α – valuation) as a graceful labeling with the additional property are an integer λ and each edge xy is either $f(x) \leq \lambda < f(y)$ or $f(y) \leq \lambda < f(x)$ [4]. It follows that such a λ must be the smaller of the two vertex labels that yield the edge labeled 1.The reported investigation ongraceful labeling of one vertex union of non-isomorphic complete bipartite graphs and one vertex union of complete bipartite graphs were reviewed for this mathematical graph modelling [5-9]. Likewise, gracefulness of a cycle with parallel P_k chords has reported with the cordial graphical Quadrilateral Snake model for the application in communication network[10, 11].Based on the graceful graphical modeling survey, this work defined simple mathematical models for the future communication applications.

THEORETICAL DEFINITIONS

Definition-1
A graph G with p vertices and q edges is said to be odd mean if there exists a function $h : V \to \{0,1,2,...,2q-1\}$ satisfying f is one –one and the induced map $h^* : E \to \{1,3,5,...2q-1\}$ defined by

$$h^*(yz) = \begin{cases} \dfrac{h(y)+h(z)}{2}, & \text{if } h(y)+h(z) \text{ is even} \\ \dfrac{h(y)+h(z)+1}{2}, & h(y)+h(z) \text{ is odd} \end{cases} \quad is \text{ a bijection.}$$

Definition-2
A function f is said to odd-even graceful labeling if $f : V(G) \to \{1,3,5,...2q+1\}$ is one to one function and $f^* : E(G) \to \{2,4,6,...2q\}$ defined by $f^*(e) = |f(u) - f(v)|$ is one to one and onto for every edge $e = (u,v) \in E(G)$. A graph G is called odd-even graceful graph if it admits an odd-even graceful labeling.

THEORETICAL MAIN RESULTS

The graph $G_1 = P_m(QS_n)$ is defined as an isomorphic Quadrilateral snake one copy gluing with each m, n is the number of Block (i.e. C_4) of Quadrilateral snake QS_n in one copy.

The graph QS_n is defined as a series connection of non-adjacent vertices with 'n' number of cycle C_4 and these vertex set V and edge set E have described using the following equations

$$V(QS_n) = \{c_k\}_{k=1}^{n+1} \cup \{u_i\}_{i=1}^{n} \cup \{v_j\}_{j=1}^{n}$$

$$E(QS_n) = \{c_k u_k\}_{k=1}^{n} \cup \{c_k v_k\}_{k=1}^{n} \cup \{u_k c_{k+1}\}_{k=1}^{n} \cup \{v_k c_{k+1}\}_{k=1}^{n}$$

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2.1. Theorem – 1

The graph $G_1 = P_m(QS_n)$, $\forall$ $m \geq 2$, $n \geq 1$ is odd-mean graph.

Proof

The graph $G_1 = P_m(QS_n)$ has $m(3n+1)$ vertices and $m(4n+1)-1$ edges. The graph $G_1 = P_m(QS_n)$ is obtained by gluing an isomorphic Quadrilateral Snake at each vertex of path P_m , $m \geq 2$ and the graph G_1 is as shown in the **Figure -2**. The odd-mean labeling for vertices of G_1 is defined as set A, B and C in equations **(1)**, **(2)** and **(3)**.

$$A = \left\{ f(c_{i,j}) \middle/ \begin{array}{l} 2(4n+1)(m-j+1)-4i+1-\dfrac{(j-1)}{2}, \quad i=1,2,3,...,n+1, \quad j=1,3,5,...,m, \ m \ is \ odd \\[3mm] (4n+1)(m-2j+4)+4i-3+\dfrac{(j-2)}{2}, \quad i=1,2,3,...,n+1, \quad j=2,4,...,m, \ m \ is \ even \end{array} \right\} \text{-- (1)}$$

$$B = \left\{ f(u_{i,j}) \middle/ \begin{array}{l} 2(4n+1)(m-j+1)-4i+\dfrac{(j-1)}{2}, \quad i=1,2,3,...,n, \quad j=1,3,5,...,m, \ m \ is \ odd \\[3mm] 2(4n+1)(m-1)+4i-5-\dfrac{(j-2)}{2}(4mn+m+1), \quad i=1,2,3,...,n, \quad j=2,4,6,...,m, \ m \ is \ even \end{array} \right\} \text{----(2)}$$

$$C = \left\{ f(v_{i,j}) \middle/ \begin{array}{l} 2(4n+1)(m-1)-4i+2-\dfrac{(j-1)}{2}(4mn+m-2), \quad i=1,2,3,...,n, j=1,3,5,...,m, \ m \ is \ odd \\[3mm] 2(4n+1)(m-2)+4i-4-m(4n+1)\dfrac{(j-2)}{2}, \quad i=1,2,3,...,n, j=2,4,6,...,m, \ m \ is \ even \end{array} \right\} \text{-- (3)}$$

From the above vertex labeling, the set A labels of the vertices form a monotonically reducing sequence for each j is odd and similar monotonically increasing sequence for each j is even was observed in the labels of the set B&C vertices. $A \cup B \cup C = \{0,1,2,...,2(m(4n+1)-1)-1\}$ and $A \cap B \cap C = \phi$. Therefore, the labels of all the vertices of G_1 are distinct. With the above labeling the corresponding edge labelings are defined by

$$A' = \left\{ f'(e_{i,j}) / \ 2(4n+1)(m-j+1)-2i-1, \ 1 \leq i \leq 4n , \ j \ is \ odd , \ 1 \leq j \leq m \right\} \text{--- (4)}$$

$$B' = \left\{ f'(e_{i,j}) / \ m(4n+1)-5+2i+m(n+1)-4(4n+1)\dfrac{(j-2)}{2}, \ 1 \leq i \leq 2n , \ j \ is \ even , \ 2 \leq j \leq m \right\} \text{--- (5)}$$

$$C' = \left\{ f'(e_{i,j}) / \ 2(4n+1)(m-j+2)-3mn-8n+2i-5, \ 2n < i \leq 4n , \ j \ is \ even, \ 2 \leq j \leq m \right\} \text{--- (6)}$$

$$D' = \left\{ f'(e_k) / \ 2[(4n+1)(m-k+1)-4n-1]-1 , \ 1 \leq k \leq m-1 \right\} \text{-- (7)}$$

the values in the sets A', B', C' and D' are all distinct and these functions explained as follows in equations (8) & (9),

$$A' \cup B' \cup C' \cup D' = \{1,3,5,...,2[m(4n+1)-1]-1\} \text{------------ (8)}$$

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$$A^{'} \cap B^{'} \cap C^{'} \cap D^{'} = \phi$$

--------------- (9)

From the above assignment, the graph labeling of vertices and edges are distinct Hence, the graph $G_1 = P_m(QS_n)$ is odd-mean graph.Illustration of the labeling has shown in the proof of **Theorem –1** and presented in **Figure.3**.

2.2. Theorem-2

The graph $G_2 = S_n + \overline{K_t}, n \geq 4, \forall t$ is odd-even graceful graph.

Proof

The graph $G_2 = S_n + \overline{K_t}$ has n+t vertices and n(t+1)-1 edges.

Odd- even graceful labeling of vertices of $S_n + \overline{K_t}$ defined by

$$f(u_i) = 2i - 1, 1 \leq i \leq n$$
$$f(u_0) = 2[n(t+1) - 1] + 1, i = 0$$
$$f(v_j) = 2nj - 1, 1 \leq j \leq t$$

Corresponding edge labelings are defined by

$$f^{'}(e_k) = 2[n(t+1) - 1] - 2(k-1), 1 \leq k \leq n-1$$
$$f^{'}(e_{i,j}) = 2(n(t+1)-1) - 2(n-1) - 2i - 8(j-1), \quad i = 0, \qquad 1 \leq j \leq t$$
$$f^{'}(e_{i,j}) = 2(n(t+1)-1) - 2(n-1) - 2i + 8(j-5), \quad 1 \leq i \leq n-1, \qquad 1 \leq j \leq t$$

From the above assignment, the labeling of vertices and edges are distinct.Hence the graph $S_n + \overline{K_t}$ is odd-even graceful graph.

Example: Figure-5. Odd-even graceful labeling of $S_4 + \overline{K_5}$

2.3. Theorem-3

The Coconut Tree $G_3 = CT(m,n)$ is odd-even graceful graph for all positive integers n and $m \geq 2$.

Proof

Let $G_3 = CT(m,n)$ be the Coconut Tree

Let $u_1, u_2, u_3, \ldots u_m, v_1, v_2, v_3, \ldots v_n$ be the vertices of Coconut Tree.

Then the vertex set $V(G) = \{ u_1, u_2, u_3, \ldots u_m, v_1, v_2, v_3, \ldots v_n \}$

The graph G_3 is as shown in the **Figure -6.**

Let $u_1, u_2, u_3, \ldots u_m$ be the vertices of the path P_m and $v_1, v_2, v_3, \ldots v_n$ be the pendant vertices attached with the end vertex of the path P_m.

Now the edge set

$E(G) = \{u_i u_{i+1} / 1 \leq i \leq m-1\} \cup \{u_m v_j / 1 \leq j \leq n\}$ then we note that $|V(CT(m,n)| = m+n$ and $m+n-1$ edges.

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$$f(u_i) = \begin{cases} 2(m+n)-i, 1 \le i \le m & i \text{ is odd} \\ i-1, 1 \le i \le m & i \text{ is even} \end{cases}$$

$$f(v_j) = m - 2 + 2j, 1 \le j \le n$$

Corresponding edge labelings are defined by

$$f^{'}(e_i) = 2(m+n-i), 1 \le i \le m-1$$

$$f^{'}(e_k) = 2n - 2k + 2 \quad , 1 \le k \le n$$

From the above assignment, vertices and edges are distinct. Hence, the graph $G_3 = CT(m,n)$ is odd-even graceful graph.

The following is an illustration of the labeling as shown in the proof of **Theorem -3**

2.4. Theorem-4

The jellyfish graph $G_4 = J(m,n)$, $m \ge 2, n \ge 2, m = n$ is odd-even graceful graph.

Proof

The graph $G_4 = J(m,n)$ has m+n+4 vertices and m+n+5 edges.

The graph $G_4 = J(m,n)$ is as shown in the **Figure -8**

Odd- even graceful labeling of vertices of $G_4 = J(m,n)$ defined by

$$f(u_0) = 2m + 2n + 11$$

$$f(u_1) = 1$$

$$f(c_0) = 3$$

$$f(c_0^{'}) = 5$$

$$f(c_i) = 2i + 7, 1 \le i \le m$$

$$f(c_j^{'}) = 2m + 2j + 9, 1 \le j \le n$$

$$f(u_0 u_1) = 2(m + n + 5)$$

$$f(u_0 c_0) = 2m + 2n + 8$$

$$f(c_0 u_1) = 2$$

$$f(u_1 c_0^{'}) = 4$$

$$f(u_0 c_0^{'}) = 2m + 2n + 6$$

Corresponding edge labelings are defined by

$$f^{'}(e_i) = 2i + 4, 1 \le i \le m$$

$$f^{'}(e_j^{'}) = 2m + 2j + 4, 1 \le j \le n$$

From the above assignment, vertices and edges are distinct. Hence, the graph $G_4 = J(m,n)$ is odd-even graceful graph.

The following is an illustration of the labeling as shown in the proof of **Theorem -4**

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CONCLUSION

Graph labeling serves as a frontier between number theory and structure of graphs.The derived Quadrilateral snake gluing of path graph and star graph can be served as useful models for communication network and also can be used for designing the drilling machines.

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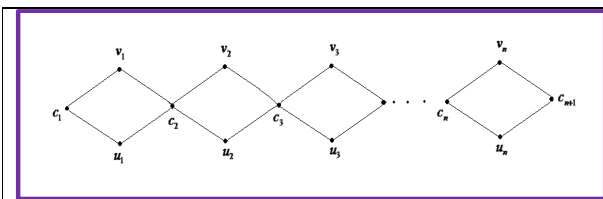
Figure -1.Quadrilateral Snake graph QS_n	**Figure- 2. The graph** $P_m(QS_n)$
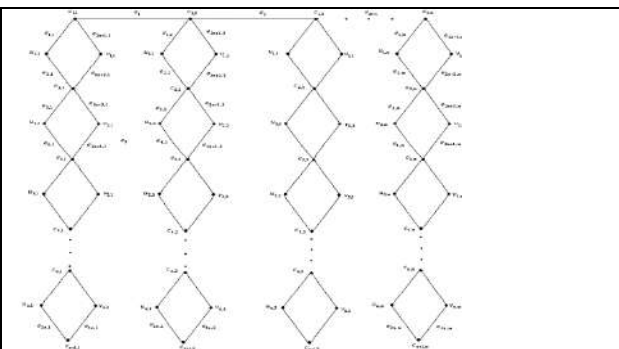
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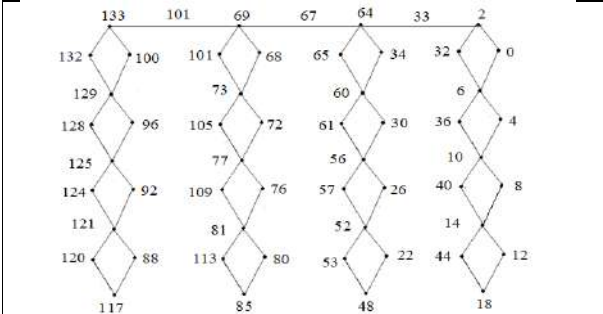
Figure- 3.Odd mean labeling of $P_4\ (QS_4\ )$
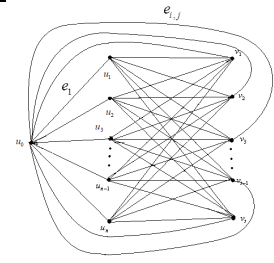
Figure -4. The graph $S_n + \overline{K}_t$
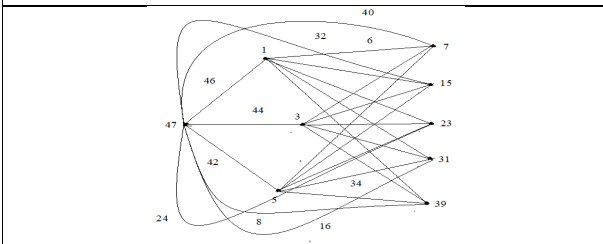
Figure-5. Odd-even graceful labeling of $S_4 + \overline{K}_5$
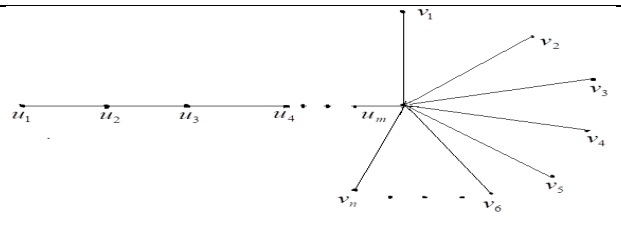
Figure-6-The graph $G_3 = CT(m,n)$
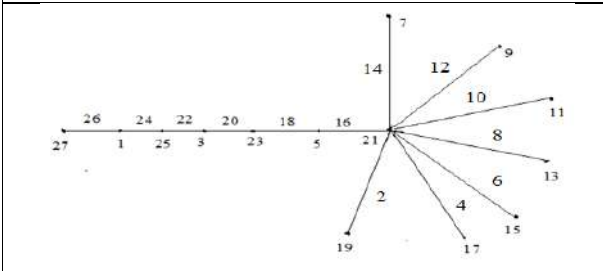
Figure-7-Odd-even graceful labeling of $CT\ (7,7)$
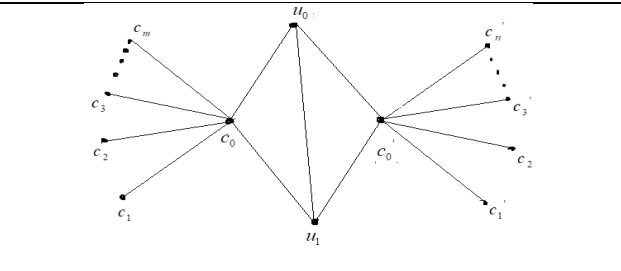
Figure-8-The graph $G_4 = J(m,n)$
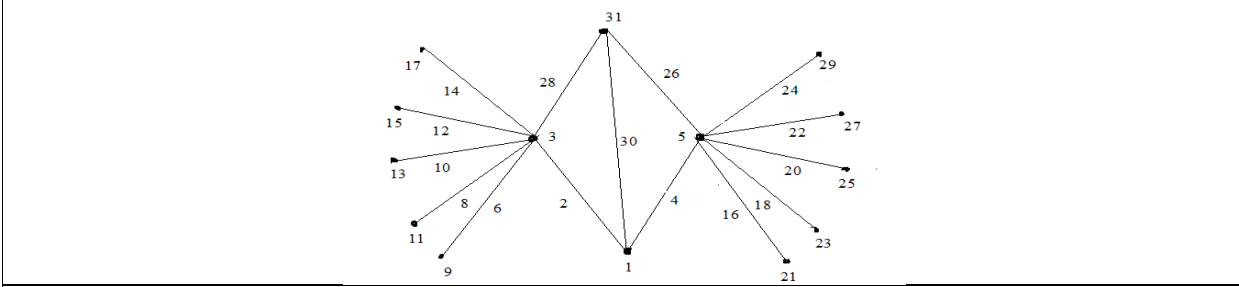
Figure-9-Odd-even graceful labeling of $J\ (5,5)$

RESEARCH ARTICLE

Conventional Neural Network Algorithm for Color Conversion in Strawberry Plant Leaf Disease Detection

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ABSTRACT

Diseases are major constraints in strawberry production throughout the word. The plant leaf diseases are mainly affected by fungi, bacteria and viruses. The leaf disease is completely destroying the quality and quantity of the leaves. Common strawberry leaf disease Leaf spot and Leaf blight. Leaf spot is a common disease, the plants cannot be cured. But if the disease is detected early, we can control the fungicides. Leaf blight is one of the most common disease in world cultivations. The leaf blight and spot images are initially a RGB images, then it is converted to HSV images because the RGB is used for color generation and color descriptor. The next step is plane separation and then performed the color features. By using Conventional Neural Network algorithm detection of leaf disease is done for detecting and calculating the affected leaf parts of the leaves and validating the disease affected area. Finally testing and training the diseased leaf images by using in Matlab R2018a

Keywords: Strawberry Plant Leaves, Leaf Blight, Leaf Spot, Conventional Neural Network algorithm

INTRODUCTION

Plant leaf disease affects the growth of their respective class; therefore, their early identification is very important. Strawberry plants are one of the most important crops of world. The cultivation is very easy to produce the quantity of the plants. The total area under strawberry is about eight million hectares. Leaf spot and Leaf blight disease is the major disease of strawberry leaves. It occurs in every country and it includes in India. Many machine learning models have been used for the detection and classification of Leaf spot and Leaf blight diseases, but after the

advancements of technology, the deep learning algorithms have great potential in terms of increased accuracy. They are many methods and algorithms that are being implemented on different plants for different diseases. Three steps that are followed by many authors.

Image preprocessing: At initial stage raw images is taken as input and smoothen through various techniques like thresholding, gray scale conversion, RGB to HIS, various filters like median filter, CIBAL color model to remove the noise and many more smoothing techniques.

Image Segmentation: The disease affected area is segmented from the leaf. For segmentation various methods are used such as k-nearest neighbor method, triangle threshold method and simple threshold methods. Disease classification: For disease classification numerous algorithms are available. SVM is most commonly used algorithm followed by artificial neural network, Genetic programming, Histogram matching method is also used with edge detection. One of the most drawbacks that found in SVM that it deals with binary systems and can classify only two inputs. Histogram matching methods gives result with poor accuracy. The common symptoms of strawberry leaf spot and Leaf blight that both appears initially a small deep purple color and its round to irregularly shaped spots and mainly appears on the upper leaf surfaces. These diseases are nearly complete defoliation and yield loss up to 50 percent or more depends upon disease severity. The leaf spot disease epidemics are affected by weather parameters such as hot and wet conditions and these diseases is a common fungal disease of strawberry plants. Losses due to Leaf spot and Leaf blight disease may go upto 15-20%. Under field conditions, initial symptoms of late leaf spot were noticed at 45-50 days after sowing of strawberry plant crops.

Leaf Spot Diseases

It's a common leaf spot of strawberry also known as Mycosphaerella leaf spot, Ramularia leaf spot, strawberry leaf spot, bird's-eye spot, gray spotness, and white spot. It is also a common fungal leaf disease that affects both wild and cultivated strawberries throughout the world. Common leaf spot was once the most economically important strawberry disease, but the use of resistant strawberry varieties/cultivars and improvements in methods for growing strawberries have been effective in managing the disease and reducing its impact. Today, the disease is often a cosmetic problem and typically has little impact on yield or fruit quality [5].

Leaf Blight Diseases

Its otherwise called as Phomopsis leaf blight. The young leaves are the Phomopsis leaf blight. The disease can also weaken older leaves in perennial plantings resulting in reduced yields the following year. In the southeast during nursery production, severe defoliation may occur and daughter plants may wilt and die due to infection of runners with symptoms that are often confused with anthracnose [4].

Disease Identification and Cycle

The leaf spot and leaf blight are the common diseases that spread among the other diseases. But in leaf spot several different fungi and one bacterium cause leaf spot diseases of strawberry. Generally, most of these leaf spots have not been of significant concern in the strawberry industry and are considered of minor importance. Losses associated with most of these diseases are rarely observed. Leaf blight are the diseases that reduces the yields. The pathogen probably derives from other plant species growing nearby strawberry nursery fields. In perennial fruit production systems, the leaf blight pathogen may overwinter in lesions on plants with infected leaves, while in annual systems the fungus is typically introduced on transplants or tips. Disease generally occurs in late summer or fall and is favored by wet weather. Primary infection occurs by rain-splashed spores, which are not airborne.

Symptoms of the Leaf Spot

1. The leaf spot first appears as circular, deep purple spots on the upper leaf surface
2. Enlarge and the centers turn grayish to white on older leaves and light brown on young leaves
3. A definite reddish purple to rusty brown border surrounds the spots.

Symptoms of Leaf Blight

1. Begins as one to several circular reddish-purple spots on the leaflet.
2. Spots enlarge to V-shaped lesions with a light brown inner zone and dark brown outer zone

Proposed Methodology

Color Image Segmentation

Color image segmentation that is based on the color feature of image pixels assumes that homogeneous colors in the image correspond to separate clusters and hence meaningful objects in the image [1]. Each cluster defines a class of pixels that share similar color properties. In this work, a segmentation of color images is tested with RGB and HSV color spaces. The HSV color space gives the best result compare to other color spaces. The median filter is used to remove the noise from the images. This filter is used to calculate the pixel values from the window into numerical order, and then replacing the pixel being considered with the middle pixel value.

Color Space Models

Based on the absorption characteristics of the human eye, colors are seen as variable combination of red, green and blue. The main purpose of the color model is to facilitate the specification of colors in some standard generally accepted way. A color model is a specification of a coordinate system and a subspace within that system where each color is represented by a single point. In order to make a detailed analysis of the leaves, the following steps are performed and the codes are written in Matlab R2018a Version.

Algorithm

Step 1: Read the image
Step 2: Convert into RGB to HSV
Step 3: Set Image h = hsv(:,1); s = hsv(:,2); v = hsv(:,3);
Step 4: Finds location of black and white pixels
Step 5: Gets the number of all pixels for each color bin
Step 6: To find the number of pixels
Step 7: Plots histogram

RGB Color Model

In this model, each color appears in its primary spectral components of red, green and blue. This model is based on Cartesian coordinate system. All values of R, G and B are assumed to be the range. The number of bites used to represent each pixel in RGB space is called the pixel depth.

HSV Color Conversion

The HSV or HSB, model describes color in terms of hue, saturation, and value (brightness). Hue corresponds directly to the concept of hue in the Color Basics section. The advantages of using hue are, the relationship between tones around the color circle is easily identified.

$$H = cos^{-1} \left[(R\text{-}G) + (G\text{-}B) / \sqrt[2]{(R-G)^2} + (R-B)(G-B) \right.$$
$$S = 1 - \frac{\min (R,G,B)}{V} \quad V = \frac{(R+G+B)}{3}$$

Conventional Neural Network Algorithm

The conventional neural network algorithm is one of the machine learning models. CNN, have been applied in different ways, including image classification, object detection, and semantic image segmentation. It has a high-level feature for image classification and perform better than another algorithm. This makes it easy for CNN to capture important feature relationships in an image and reduces the number of parameters.CNN algorithms perform both the 2-dimensional images and 3-dimentional images. Here, we using this algorithm validating the leaf spot and leaf

blight images after the color conversion. Finally, the testing and training the leaf spot and leaf blight images. The whole process is divided in to three phases.

1. The input images are collected from the Kaggle website and upload the images.
2. Image segmentation pre-processing the images that includes process of image segmentation, image enhancement by using RGB to HSV color conversion. Then convert each image into an array and each image name is convert into a binary field.
3. CNN classifiers are trained to identify diseases in leaf spot and leaf blight.

EXPERIMENTAL ANALYSIS AND RESULT

All experiments are tested with MATLAB R2018a. For input data disease, a sample of strawberry plant leaves with leaf spot and **late blight images are considered. The data set contains 200** leaf spot and **late blight images. The affected disease part of the images is calculated and finds the accuracy based on** Conventional Neural Network**. The time and accuracy are the two main concepts in** Conventional Neural Network algorithm **for calculating the leafdisease detection.** The result can be for leaf spot images by using Conventional Neural Network algorithm it gives 85% and the leaf blight is 83%.

CONCLUSION

In this paper we have proposed an image-processing based approach to automatically classify the diseased leaves of leaf spot and late blight diseases in strawberry plant leaf images and also provide the cure for the same which would be beneficial to beginners in farming or gardening. In our approach we have color features image segmentation of a leaf to train the affected images with region based conventional neural network algorithm which is more efficient than the traditional support vector machine algorithm to segment the test images. For future we can expand it to other plant leaf images.

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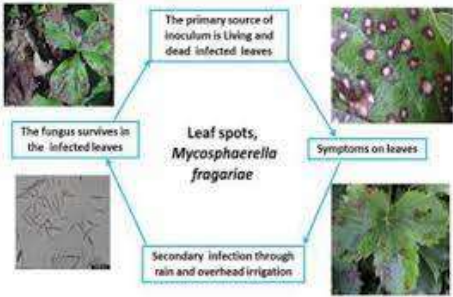
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Fig 1: Healthy Leaf

Fig 2: Infected Leaf

Fig 3: Infected Leaf

Fig 4: Life Cycle of the Leaf Spot disease in Strawberry Plant

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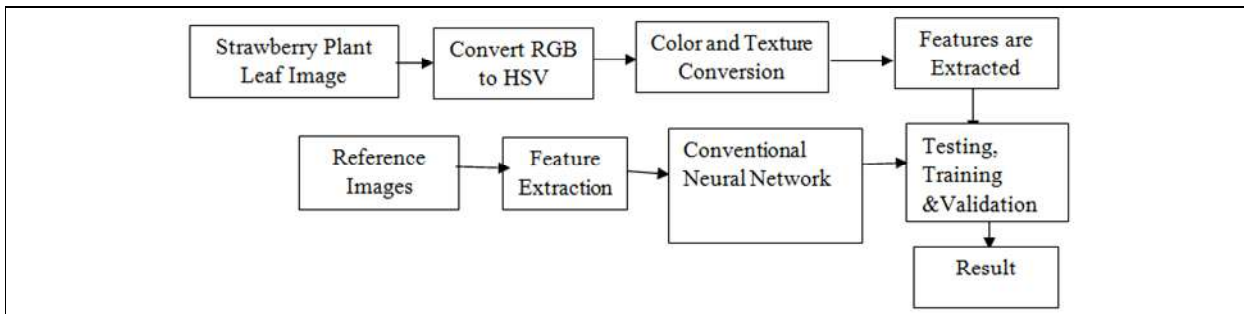
Fig 5: Block Diagram
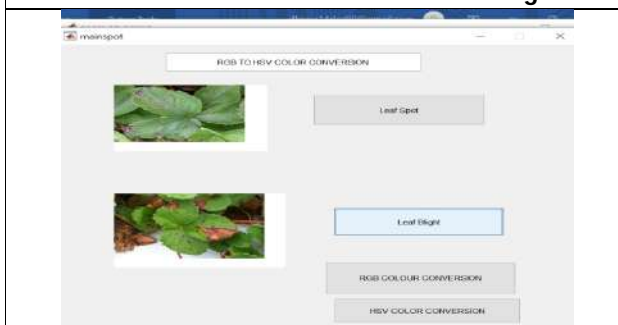
Fig 6: Input Image
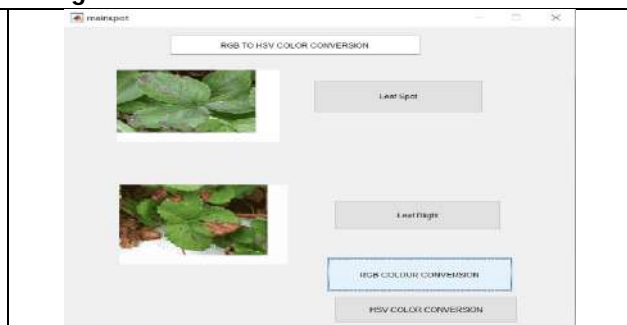
Fig 7: RGB color Conversion
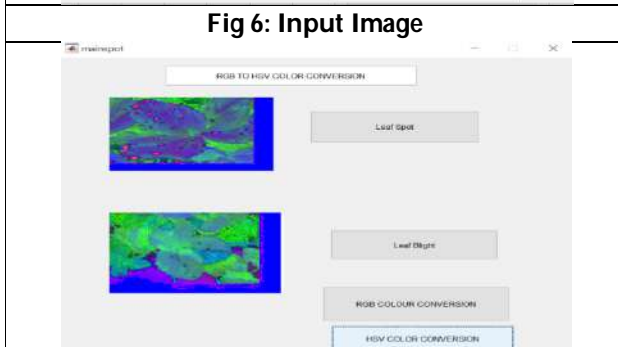
Fig 8: RGB to HSV Color Conversion
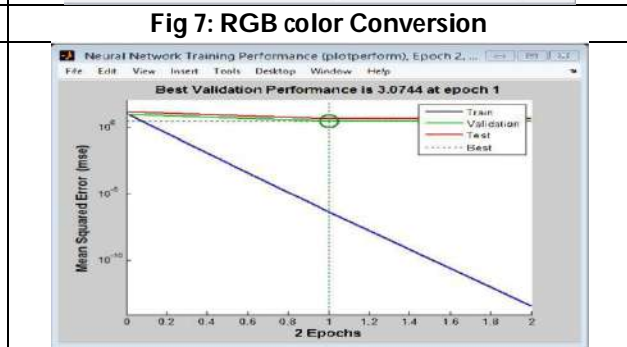
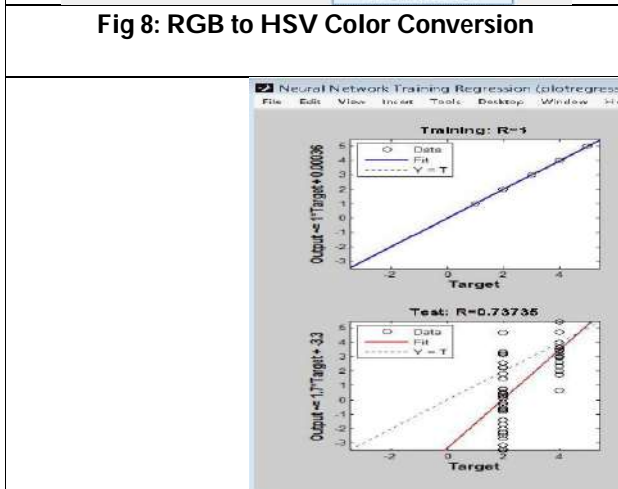**Fig 10: Performance Analysis for Leaf Spot and Late Blight**

Fig 11: Validation, Testing and Training the Strawberry Plant Leaf Images

RESEARCH ARTICLE

Role of Digital Technology in Agriculture

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ABSTRACT

Digital agriculture involves the application of digital technology to integrate agricultural output from the paddock to the consumer. Digital innovations can assist developing countries for overcoming global poverty and hunger quicker in rural areas. Indoor vertical farming, automation and robotics, livestock technology, modern greenhouse practices, precision agriculture and artificial intelligence and blockchain are major technological advancements. Farm equipment is connected to software platforms that capture on farm data and enable studies of soil and climate conditions in specific location to provide farmers with guidance regarding seed choice and more precise application of pesticides and fertilizers. Mobile phones have one of the highest adoption rates of any technology developed within the last century. Digitalisation will bring consumers and farmers closer together. The extensive information available on plants and animals makes farming more transparent to consumers. In the long run smart farming will affect agricultural production.

Keywords: Digital agriculture, innovations, vertical farming, Mobile phones, Digitalisation

INTRODUCTION

The world is being increasingly connected as a result of digital technology. At smaller, faster, cheaper and more efficient way, mobile devices are being used. Many businesses and farmers are receiving assistance in making more informed decisions. The farmers are being assisted in using more accurate amounts of water, fertilizers and maintain better control over their operations. Digital technologies make easy many processes such as planning farming activities, budgeting, reporting and monitoring on multiple tasks and performances. Digital technology is used in farm machinery, livestock handling facilities, agronomy, communication and other areas [1]. Agriculture is at the

start of a new revolution, one in which data and connectivity will play a key role. Artificial intelligence, analytics, connected sensors, and other advanced technologies have the potential to increase yields, enhance the efficiency of water and other inputs. These can help for building sustainability and resilience across crop cultivation and animal husbandry [2].Digital farming technologies include the application of sensors, automation, and robots in production systems ([3].; [4].).

Digital Agriculture-Requirement of the present time

Remote sensing data on soil conditions can help farmers manage their crop [5]. Mobile phones lower the cost of information and improve farmers' access to markets and financial assistance [6]. Entrepreneurs in Africa are interested in how farmers operate and how they may increase output. The barrier of entry into farming technology has decreased as cloud computing, computing systems, connectivity, open-source software, and other digital tools are more affordable and accessible. Farmers, investors and entrepreneurs can use digital technology to improve efficiency of food production and consumption in Africa. From precision farming to a more efficient food supply chain, technology has the potential to bring major economic, social and environmental benefits [7].Digital agriculture involves the application of digital technology to integrate agricultural production from the paddock to the costumer. Agricultural industries can benefit from these technologies by more tools and information to make better decisions and increase productivity [8]. In rural areas of developing countries where majority of people rely on agriculture for their living, digital technology can assist for overcoming global poverty and hunger more quickly. In digital agriculture, farmers use mobile phones and other technologies that could revolutionise for the communities to secure and improve their livelihoods [9].It is required more productive, efficient, sustainable, inclusive, transparent and resilient food systems for achieving the UN Sustainable Development Goal of a 'world with zero hunger' by 2030 [10].

Digital skills and people who are capable of using digital devices, comprehending outputs and developing programmes and applications are in high demand as a result of digitalisation. Basic literacy, numeracy, data handling and communication skills are required. Education must improve quickly in areas where skills are low [11]. Indoor vertical farming, automation and robotics, livestock technology, modern greenhouse practices, precision agriculture and artificial intelligence and blockchain have been all major technological advancements in the space. Indoor vertical farming has the potential to increase crop yields, overcome land constraints and shorten supply chain distances. Vertical farms use up to 70% less water than traditional farms [12]. Mobile phones are at the top of the list of digital technologies revolutionising agriculture. They have the highest rates of adoption of any technology developed within the last century. As of 2019, Almost 5.2 billion people- two thirds of the world's populations were current active users of mobile phones. Mobile internet is also on the rise, with 49 per cent of the world's population having utilised internet services on a mobile device. Only 9 per cent of the world's population lives outside of a mobile network's coverage region. Many rural people's lives including the small scale farmers have been revolutionised by mobile phones. Farmers may access solutions, such as advisory services that offer ideas on raising outputs, keeping livestock healthy and up-to-date weather information even if they do not have access to mobile internet. Farmers may even access wider choice of high-tech solutions via smart phone applications which can diagnose ailing crops and recommend remedies for many types of pests, diseases and nutrient deficiencies [13].

Precision agriculture technologies are transforming the face of modern farming. Digital advancements such as wireless communication, data analytics, and data-driven genome editing, are rapidly being applied in agriculture as they provide more accuracy in decision making and practice. Farm equipment is connected to software platforms that track on-farm data and enable analysis of soil and climate conditions. Agricultural genome makes use of big data generated from computer assisted genomic mapping to determine edits to the DNA of living organisms which promise a more accurate way to modify a plant's genetic code to express new traits for improving crop production [14]. Consumers and farmers will be closer together due to digitalisation. Consumers can learn about plants and animals that will make farming more transparent. In the long run smart farming will affect agricultural production [15].

Initiatives

The Government of India in 2015 announced the Digital India Programme which aims to transform India's public service sector into the digital space. Agriculture is the largest employment vertical with a GDP share of 14% plays an important role in this digital effort. Due to mechanisation and knowledge dissemination programmes, India has witnessed a substantial increase in yields and crop diversification. But consumption is increasing due to population explosion and rising income. The Indian Government has launched a number of initiatives. IIT Kharagpur is educating the farming community through training and putting on outreach events [16].Researchers at MIT have found a promising method for protecting seeds from water shortage stress during their crucial germination phase and additionally providing the plants with more nutrition at the same time [17]. Researchers at the Singapore-MIT Alliance for Research and Technology (SMART) and Temasek Life Sciences Laboratory (TLL) have designed a portable optical sensor which can reveal whether a plant is under stress. This device helps as a new tool for early analysis and real time monitoring of plant health in field conditions [18].

CONCLUSION

Agriculture is made up of a series of complex interconnected processes. It should be organised into efficient stages to ensure a good yield. A triangle formed by the farmer, technology and the service and consulting concept will determine the success of a given product [19]. Agriculture like other areas in the economy will be digitised in the future. Government should spend time and financial resources for socialising the digitisation benefits [20].Inadequatet connectivity in rural areas, high service charges, and a lack of basic computer literacy and understanding are obstacles for the quick development of e-agriculture. Physical infrastructure, power, broadband and transportation all require substantial investment [21].

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RESEARCH ARTICLE

(1,2)*- $\breve{g}_\alpha$- Closed Sets and It's Properties

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ABSTRACT

We introduce a new classes of sets namely (1,2)*-$\breve{g}_\alpha$-closed sets, (1,2)*-$\Lambda_\mathcal{G}$-set and (1,2)*-$\lambda_\mathcal{G}$-set are study in bitopological spaces. We prove that this classes lies between (1,2)*-α-closed sets and (1,2)*-αg-closed sets. Also we discuss some essential properties of (1,2)*-$\breve{g}_\alpha$-closed sets in present of this paper.

INTRODUCTION

The perceptions of bitopological spaces is to introduced and studied by J. C. Kelly [2]. Recently, More generalizations of closed sets and it is properties were introduced and investigated by various researchers for some example ([6, 5, 9]) and so on. We introduce a new classes of sets namely (1,2)*-$\breve{g}_\alpha$-closed sets, (1,2)*-$\Lambda_\mathcal{G}$-set and (1,2)*-$\lambda_\mathcal{G}$-set are study in bitopological spaces. We prove that this classes lies between (1,2)*-α-closed sets and (1,2)*-αg-closed sets. Also we discuss some essential properties of (1,2)*-$\breve{g}_\alpha$-closed sets in present of this paper.

Preliminaries

Throughout this paper (X, τ_1, τ_2) and (Y, σ_1, σ_2) (or simply X and Y) represents the non-empty bitopological spaces on which no separation axiom are assumed, unless otherwise mentioned. For a subset A of X, $\tau_{1,2}$-$cl(A)$ and $\tau_{1,2}$-$int(A)$ represents the closure of A and interior of A respectively.

Definition 2.1 [3] A subset A of a bitopological space (X, τ_1, τ_2) or X is called
1. a (1,2)*-semi open set if $A \subseteq \tau_{1,2}$-$cl(\tau_{1,2}$-$int(A))$.
2. a (1,2)*-pre open set if $A \subseteq \tau_{1,2}$-$int(\tau_{1,2}$-$cl(A))$.

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3. a $(1,2)^\star$-α-open set if $A \subseteq \tau_{1,2}\text{-}int(\tau_{1,2}\text{-}cl(\tau_{1,2}\text{-}int(A)))$.
4. a $(1,2)^\star$-β-open (or) a $(1,2)^\star$-semi-pre open set [8] if $A \subseteq \tau_{1,2}\text{-}cl(\tau_{1,2}\text{-}int(\tau_{1,2}\text{-}cl(A)))$.
The complements of the above mentioned sets are called their respective closed sets.

Definition 2.2 A subset A of a bitopological space (X, τ_1, τ_2) *or X is said to be*
1. a $(1,2)^\star$-generalized closed set (briefly, $(1,2)^\star$-g-closed) [9] if $(1,2)^\star$-$cl(A) \subseteq U$ whenever $A \subseteq U$ and U is $\tau_{1,2}$-open.
2. a $(1,2)^\star$-semi generalized closed set (briefly, $(1,2)^\star$-sg-closed) [7] if $(1,2)^\star$-$scl(A) \subseteq U$ whenever $A \subseteq U$ and U is $(1,2)^\star$-semi-open.
3. a $(1,2)^\star$-generalized semi-closed (briefly, $(1,2)^\star$-gs-closed) set [1] if $(1,2)^\star$-$scl(A) \subseteq U$ whenever $A \subseteq U$ and U is $\tau_{1,2}$-open.
4. an $(1,2)^\star$-α-generalized closed (briefly, $(1,2)^\star$-αg-closed) set [4] if $(1,2)^\star$-$\alpha cl(A) \subseteq U$ whenever $A \subseteq U$ and U is $\tau_{1,2}$-open.
5. a $(1,2)^\star$-generalized semi-preclosed (briefly, $(1,2)^\star$-gsp-closed) set [4] if $(1,2)^\star$-$\beta cl(A) \subseteq U$ whenever $A \subseteq U$ and U is $\tau_{1,2}$-open.
6. a $(1,2)^\star$-$\hat{g}$-closed set [6] if $\tau_{1,2}$-$cl(A) \subseteq U$ whenever $A \subseteq U$ and U is $(1,2)^\star$-sg-open.
7. a $(1,2)^\star$-$\hat{g}_1$-closed set [5] if $\tau_{1,2}$-$cl(A) \subseteq U$ whenever $A \subseteq U$ and U is $(1,2)^\star$-$\hat{g}$-open.
8. a $(1,2)^\star$-$\mathcal{G}$-closed set [5] if $(1,2)^\star$-$scl(A) \subseteq U$ whenever $A \subseteq U$ and U is $(1,2)^\star$-$\hat{g}_1$-open.
9. a $(1,2)^\star$-$\breve{g}$-closed set [5] if $\tau_{1,2}$-$cl(A) \subseteq U$ whenever $A \subseteq U$ and U is $(1,2)^\star$-$\mathcal{G}$-open.
The complements of the above mentioned closed sets are called their respective open sets.

ON $(1,2)^\star$-$\breve{g}_\alpha$-CLOSED SETS
Definition 3.1 *A subset A of a space (X, τ_1, τ_2) is said to be an $(1,2)^\star$-$\breve{g}_\alpha$-closed set if $\tau_{1,2}$-$\alpha cl(A) \subseteq U$ whenever $A \subseteq U$ and U is $(1,2)^\star$-$\mathcal{G}$-open.* The complement of $(1,2)^\star$-$\breve{g}_\alpha$-closed set is called $(1,2)^\star$-$\breve{g}_\alpha$-open set.
The collection of all $(1,2)^\star$-$\breve{g}_\alpha$-closed (resp. $(1,2)^\star$-$\breve{g}_\alpha$-open) sets in (X, τ_1, τ_2) is denoted by $(1,2)^\star$-$\breve{g}_\alpha C(X)$ (resp. $(1,2)^\star$-$\breve{g}_\alpha O(X)$).

Proposition 3.2 *In a space (X, τ_1, τ_2), each $(1,2)^\star$-α-closed set is $(1,2)^\star$-$\breve{g}_\alpha$-closed.*
Proof. Let A be an $(1,2)^\star$-α-closed set and U be any $(1,2)^\star$-$\mathcal{G}$-open set containing A. Since A is $(1,2)^\star$-α-closed, we have $\tau_{1,2}$-$\alpha cl(A) = A \subseteq U$. Thus A is $(1,2)^\star$-$\breve{g}_\alpha$-closed.

Remark 3.3 The converse of Proposition 3.2 need not be true as seen from the following Example.

Example 3.4 *Let $X = \{a, b, c\}$ with $\tau_1 = \{\Phi, \{a, b\}, X\}$ and $\tau_2 = \{\Phi, X\}$ then $\tau_{1,2} = \{\Phi, \{a, b\}, X\}$. In the space X, then $(1,2)^\star$-$\breve{g}_\alpha C(X) = \{\Phi, \{c\}, \{a, c\}, \{b, c\}, X\}$ and $(1,2)^\star$-$\alpha C(X) = \{\Phi, \{c\}, X\}$. We have the subset $\{a, c\}$ is $(1,2)^\star$-$\breve{g}_\alpha$-closed set but not $(1,2)^\star$-α-closed.*

Proposition 3.5 *In a space (X, τ_1, τ_2), each $(1,2)^\star$-$\breve{g}$-closed set is $(1,2)^\star$-$\breve{g}_\alpha$-closed.*
Proof. Let A be a $(1,2)^\star$-$\breve{g}$-closed set and U be any $(1,2)^\star$-$\mathcal{G}$-open set containing A. Since A is $(1,2)^\star$-$\breve{g}$-closed, we have $U \supseteq cl(A) \supseteq \tau_{1,2}$-$\alpha cl(A)$. Hence A is $(1,2)^\star$-$\breve{g}_\alpha$-closed.

Remark 3.6 The converse of Proposition 3.5 need not be true as seen from the following Example.

Example 3.7 *Let $X = \{a, b, c\}$ with $\tau_1 = \{\Phi, X\}$ and $\tau_2 = \{\Phi, \{b\}, X\}$ then $\tau_{1,2} = \{\Phi, \{b\}, X\}$. In a space X, then $(1,2)^\star$-$\breve{g}_\alpha C(X) = \{\Phi, \{a, c\}, X\}$ and $(1,2)^\star$-$\breve{g}_\alpha C(X) = \{\Phi, \{a\}, \{c\}, \{a, c\}, X\}$. We have the subset $\{a\}$ is $(1,2)^\star$-$\breve{g}_\alpha$-closed set but not $(1,2)^\star$-$\breve{g}$-closed.*

Proposition 3.8 *In a space (X, τ_1, τ_2), each $(1,2)^\star$-$\breve{g}_\alpha$-closed set is $(1,2)^\star$-αg-closed.*
Proof. Let A be an $(1,2)^\star$-$\breve{g}_\alpha$-closed set and U be any $\tau_{1,2}$-open set containing A. Since any $\tau_{1,2}$-open set is $(1,2)^\star$-$\mathcal{G}$-open, then $\tau_{1,2}$-$\alpha cl(A) \subseteq U$. Thus A is $(1,2)^\star$-αg-closed.

Remark 3.9 The converse of Proposition 3.8 need not be true as seen from the following Example.

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Example 3.10 Let $X = \{a, b, c\}$ with $\tau_1 = \{\Phi, \{c\}, X\}$ and $\tau_2 = \{\Phi, X\}$ then $\tau_{1,2} = \{\Phi, \{c\}, X\}$. Then $(1,2)^*\text{-}ǧ_\alpha C(X) = \{\Phi, \{a\}, \{b\}, \{a, b\}, X\}$ and $(1,2)^*\text{-}\alpha g C(X) = \{\Phi, , \{a\}, \{b\}, \{a, b\}, \{a, c\}, \{b, c\}, X\}$. In a space X, we have the subset $\{a, c\}$ *is* $(1,2)^*\text{-}\alpha g$ set but not $(1,2)^*\text{-}ǧ_\alpha\text{-}closed.$

Proposition 3.11 In a space (X, τ_1, τ_2), each $(1,2)^*\text{-}ǧ_\alpha$-closed set is $(1,2)^*\text{-}gs\text{-}closed$ $((1,2)^*\text{-}sg\text{-}closed)$.
Proof. Let A be an $(1,2)^*\text{-}ǧ_\alpha$-closed set and U be any $\tau_{1,2}$-open set $((1,2)^*$-semi-open set) containing A. Since any $\tau_{1,2}$-open $((1,2)^*$-semi-open) set is $(1,2)^*\text{-}\mathcal{G}$-open, then $\tau_{1,2}\text{-}scl(A) \subseteq \tau_{1,2}\text{-}\alpha cl(A) \subseteq A$. Thus A is $(1,2)^*\text{-}gs$-closed $((1,2)^*\text{-}sg$-closed).

Remark 3.12 The converse of Proposition 3.11 need not be true as seen from the following Example.

Example 3.13 *Let* $X = \{a, b, c\}$ *with* $\tau_1 = \{\Phi, \{a\}, \{a, b\}, X\}$ *and* $\tau_1 = \{\Phi, \{b\}, \{a, b\}, X\}$ *then* $\tau_1 = \{\Phi, \{a\}, \{b\}, \{a, b\}, X\}$. *We have* $(1,2)^*\text{-}ǧ_\alpha C(X) = \{\phi, \{c\}, \{a, c\}, \{b, c\}, X\}$ *and* $(1,2)^*\text{-}sg C(X) = (1,2)^*\text{-}gs C(X) = \{\Phi, \{a\}, \{b\}, \{c\}, \{a, c\}, \{b, c\}, X\}$. *In the space X, then the subset* $\{a\}$ *is both* $(1,2)^*\text{-}sg$-*closed set and* $(1,2)^*\text{-}gs$-*closed set but not* $(1,2)^*\text{-}ǧ_\alpha$-*closed.*

Proposition 3.14 *In a space* (X, τ_1, τ_2), *each* $(1,2)^*\text{-}ǧ_\alpha$-*closed set is* $(1,2)^*\text{-}gsp$-*closed.*

Proof. Let A be an $(1,2)^*\text{-}ǧ_\alpha$--closed set and U be any $\tau_{1,2}$-open set containing A. Since any $\tau_{1,2}$-open set is $(1,2)^*\text{-}\mathcal{G}$-open, then $\tau_{1,2}\text{-}spcl(A) \subseteq \tau_{1,2}\text{-}cl(A) \subseteq U$. Hence A is $(1,2)^*\text{-}gsp$-closed.

Remark 3.15 The converse of Proposition 3.14 need not be true as seen from the following Example.

Example3.16 In Example 3.7, we have $(1,2)^*\text{-}gsp C(X) = \{\Phi, \{a\}, \{c\}, \{a, b\}, \{a, c\}, \{b, c\}, X\}$. In the space X, then the subset $\{a, b\}$ *is* $(1,2)^*\text{-}gsp$-closed set but not $(1,2)^*\text{-}ǧ_\alpha\text{-}closed.$

Remark 3.17 The following Examples show that $(1,2)^*\text{-}ǧ_\alpha$-closedness is independent of $(1,2)^*$-semi-closedness and $(1,2)^*\text{-}g$-closedness.
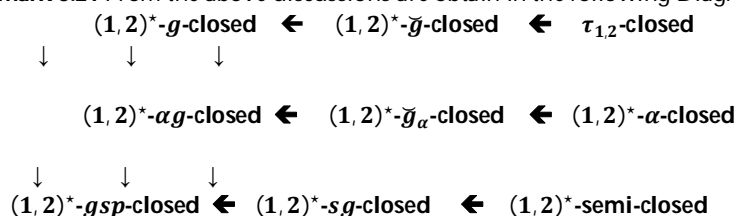
Example 3.18 In Example 3.13, we have $(1,2)^*\text{-}sC(X) = \{\Phi, \{a\}, \{b\}, \{c\}, \{a, c\}, \{b, c\}, X\}$. In the space, then the subset $\{b\}$ *is* $(1,2)^*$-semi-closed set but not $(1,2)^*\text{-}ǧ_\alpha\text{-}closed.$

Example 3.19 In Example 3.4, we have $(1,2)^*\text{-}sC(X) = \{\Phi, \{c\}, X\}$. In the space, then the subset $\{b, c\}$ *is* $(1,2)^*\text{-}ǧ_\alpha$-closed set but not $(1,2)^*$-semi-closed.

Example 3.20 Let $X = \{a, b, c\}$ with $\tau_1 = \{\Phi, \{a\}, X\}$ and $\tau_2 = \{\Phi, \{a, b\}, X\}$ then $\tau_{1,2} = \{\Phi, \{a\}, \{a, b\}, X\}$. We have $(1,2)^*\text{-}ǧ_\alpha C(X) = \{\Phi, \{b\}, \{c\}, \{b, c\}, X\}$ *and* $(1,2)^*\text{-}g C(X) = \{\Phi, \{c\}, \{a, c\}, \{b, c\}, X\}$. In the space, then
1. the subset $\{b\}$ is $(1,2)^*\text{-}ǧ_\alpha$-closed set but not $(1,2)^*\text{-}g$-closed.
2. the subset $\{a, c\}$ is $(1,2)^*\text{-}g$-closed set but not $(1,2)^*\text{-}ǧ_\alpha$-closed.

Remark 3.21 From the above discussions are obtain in the following Diagram.

$$(1,2)^*\text{-}g\text{-closed} \quad \leftarrow \quad (1,2)^*\text{-}ǧ\text{-closed} \quad \leftarrow \quad \tau_{1,2}\text{-closed}$$
$$\downarrow \qquad \downarrow \qquad \downarrow$$
$$(1,2)^*\text{-}\alpha g\text{-closed} \quad \leftarrow \quad (1,2)^*\text{-}ǧ_\alpha\text{-closed} \quad \leftarrow \quad (1,2)^*\text{-}\alpha\text{-closed}$$
$$\downarrow \qquad \downarrow \qquad \downarrow$$
$$(1,2)^*\text{-}gsp\text{-closed} \quad \leftarrow \quad (1,2)^*\text{-}sg\text{-closed} \quad \leftarrow \quad (1,2)^*\text{-semi-closed}$$

None of above implications are reversible.

Properties of $(1,2)^*$-$\breve{g}_\alpha$-Closed Sets

Definition 4.1 The intersection of all $(1,2)^*$-$\mathcal{G}$-open subsets in (X, τ_1, τ_2) containing A is said to be a $(1,2)^*$-$\mathcal{G}$-kernel of A and denoted by $(1,2)^*$-$\mathcal{G}$-$Ker(A)$.

Lemma 4.2 *A subset A of (X, τ_1, τ_2) is $(1,2)^*$-$\breve{g}_\alpha$-closed iff $\tau_{1,2}$-$\alpha cl(A) \subseteq (1,2)^*$-$\mathcal{G}$-$Ker(A)$.*
Proof. Suppose that A is $(1,2)^*$-$\breve{g}_\alpha$-closed. Then $(1,2)^*$-$\alpha cl(A) \subseteq U$ whenever $A \subseteq U$ and U is $(1,2)^*$-$\mathcal{G}$-open. Let $x \notin \tau_{1,2}$-$\alpha cl(A)$. If $x \notin (1,2)^*$-$\mathcal{G}$-$Ker(A)$,, then there is $(1,2)^*$-$\mathcal{G}$-open set U containing A such that $x \notin U$. Since U is $(1,2)^*$-$\mathcal{G}$-open set containing A, we have $x \notin \tau_{1,2}$-$\alpha cl(A)$ and this is a contradiction.
Conversely, let $\tau_{1,2}$-$\alpha cl(A) \subseteq (1,2)^*$-$\mathcal{G}$-$Ker(A)$. If U is any $(1,2)^*$-$\mathcal{G}$-open set containing A, then $\tau_{1,2}$-$\alpha cl(A) \subseteq (1,2)^*$-$\mathcal{G}$-$Ker(A) \subseteq U$. Therefore, A is $(1,2)^*$-$\breve{g}_\alpha$-closed.

Proposition 4.3 *If A and B are $(1,2)^*$-$\breve{g}_\alpha$-closed sets in (X, τ_1, τ_2), then $A \cup B$ is $(1,2)^*$-$\breve{g}_\alpha$-closed in (X, τ_1, τ_2).*
Proof. If $A \cup B \subseteq U$ and U is $(1,2)^*$-$\mathcal{G}$-open, then $A \subseteq U$ and $B \subseteq U$. Since A and B are $(1,2)^*$-$\breve{g}_\alpha$-closed, $U \supseteq \tau_{1,2}$-$\alpha cl(A)$ and $U \supseteq \tau_{1,2} - \alpha cl(B)$ and hence $U \supseteq \tau_{1,2}$-$\alpha cl(A) \cup \tau_{1,2}$-$\alpha cl(B) = \tau_{1,2}$-$\alpha cl(A \cup B)$. Thus $A \cup B$ is $(1,2)^*$-$\breve{g}_\alpha$-closed in (X, τ_1, τ_2).

Proposition 4.4 *If a set A is $(1,2)^*$-$\breve{g}_\alpha$-closed in (X, τ_1, τ_2) and $A \subseteq B \subseteq \tau_{1,2}$-$\alpha cl(A)$, then B is $(1,2)^*$-$\breve{g}_\alpha$-closed in (X, τ_1, τ_2).*
Proof. Let U be $(1,2)^*$-$\mathcal{G}$-open set in (X, τ_1, τ_2) such that $B \subseteq U$. Then $A \subseteq U$. Since A is an $(1,2)^*$-$\breve{g}_\alpha$-closed set, $\tau_{1,2}$-$\alpha cl(A) \subseteq U$. Also $\tau_{1,2}$-$\alpha cl(B) = \tau_{1,2}$-$\alpha cl(A) \subseteq U$. Hence B is also an $(1,2)^*$-$\breve{g}_\alpha$-closed in (X, τ_1, τ_2).

Proposition 4.5 *If A is $(1,2)^*$-$\mathcal{G}$-open and $(1,2)^*$-$\breve{g}_\alpha$-closed in (X, τ_1, τ_2), then A is $(1,2)^*$-α-closed in (X, τ_1, τ_2).*
Proof. Since A is $(1,2)^*$-$\mathcal{G}$-open and $(1,2)^*$-$\breve{g}_\alpha$-closed, $\tau_{1,2}$-$\alpha cl(A) \subseteq A$ and hence A is $(1,2)^*$-α-closed in (X, τ_1, τ_2).

Proposition 4.6 *For each $x \in X$, either $\{x\}$ is $(1,2)^*$-$\mathcal{G}$-closed or $\{x\}^c$ is $(1,2)^*$-$\breve{g}_\alpha$-closed in (X, τ_1, τ_2).*
Proof. Suppose that $\{x\}$ is not $(1,2)^*$-$\mathcal{G}$-closed in (X, τ_1, τ_2). Then $\{x\}^c$ is not $(1,2)^*$-$\mathcal{G}$-open and the only $(1,2)^*$-$\mathcal{G}$-open set containing $\{x\}^c$ is the space X itself. Therefore $\tau_{1,2}$-$\alpha cl(\{x\}^c) \subseteq X$ and so $\{x\}^c$ is $(1,2)^*$-$\breve{g}_\alpha$-closed in (X, τ_1, τ_2).

Definition 4.7 *A subset A of a space (X, τ_1, τ_2) is said to be $(1,2)^*$-$\Lambda_\mathcal{G}$-set if $A = (1,2)^*$-$\mathcal{G}$-$Ker(A)$.*

Definition 4.8 *A subset A of a space (X, τ_1, τ_2) is called $(1,2)^*$-$\lambda_\mathcal{G}$-closed if $A = S \cap T$ where S is a $(1,2)^*$-$\Lambda_\mathcal{G}$-set and T is $(1,2)^*$-α-closed.* The complement of $(1,2)^*$-$\lambda_\mathcal{G}$-closed set is called $(1,2)^*$-$\lambda_\mathcal{G}$-open set.
The collection of all $(1,2)^*$-$\lambda_\mathcal{G}$-closed (resp. $(1,2)^*$-$\lambda_\mathcal{G}$-open) sets in (X, τ_1, τ_2) is denoted by $(1,2)^*$-$\lambda_\mathcal{G} C(X)$ (resp. $(1,2)^*$-$\lambda_\mathcal{G} O(X)$).

Lemma 4.9 *For a subset A of a topological space (X, τ_1, τ_2), the following conditions are equivalent.*
1. *A is $(1,2)^*$-$\lambda_\mathcal{G}$-closed.*
2. *$A = S \cap \tau_{1,2}$-$\alpha cl(A)$ where S is a $(1,2)^*$-$\Lambda_\mathcal{G}$-set.*
3. *$a = (1,2)^*$-$\mathcal{G}$-$Ker(A) \cap \tau_{1,2}$-$\alpha cl(A)$.*

Lemma 4.10 *In a space (X, τ_1, τ_2),*
1. *every $(1,2)^*$-α-closed set is $(1,2)^*$-$\lambda_\mathcal{G}$-closed.*
2. *every $(1,2)^*$-$\Lambda_\mathcal{G}$-set is $(1,2)^*$-$\lambda_\mathcal{G}$-closed.*

Remark 4.11 *The converses of Lemma 4.10 need not be true as seen from the following Examples.*

Example 4.12 *Let $X = \{a, b, c, d, e\}$ with $\tau_1 = \{\Phi, \{a\}, X\}$ and $\tau_2 = \{\Phi, X\}$ then $\tau_{1,2} = \{\Phi, \{a\}, X\}$, we have*
1. $(1,2)^*$-$\alpha C(X) = \{\Phi, \{b\}, \{c\}, \{b, c\}, X\}$ and $(1,2)^*$-$\lambda_\mathcal{G} C(X) = P(X)$. In the space X, then the subset $\{a\}$ is $(1,2)^*$-$\lambda_\mathcal{G}$-closed set but not $(1,2)^*$-α-closed.

2. $(1,2)^\star$-$\Lambda_{\mathcal{G}}$-sets are $\{\Phi, \{a\}, \{a, b\}, \{a, c\}, X\}$ and $(1,2)^\star$-$\lambda_{\mathcal{G}} C(X) = P(X)$. In the space X, then the subset $\{b\}$ is $(1,2)^\star$-$\lambda_{\mathcal{G}}$-closed set but not $(1,2)^\star$-$\Lambda_{\mathcal{G}}$-set.

Theorem 4.13 *For a subset A of a topological space (X, τ_1, τ_2), the following conditions are equivalent.*
1. A is $(1,2)^\star$-α-closed.
2. A is $(1,2)^\star$-$\mathfrak{g}_{\alpha}$ and $(1,2)^\star$-$\lambda_{\mathcal{G}}$.
Proof.(1) $\Rightarrow$ (2). Obvious.
(2) $\Rightarrow$ (1). Since A is $(1,2)^\star$-$\mathfrak{g}_{\alpha}$-closed, so by Lemma 4.2, $\tau_{1,2}$-$\alpha cl(A) \subseteq (1,2)^\star$-$\mathcal{G}$-$Ker(A)$. Since A is $(1,2)^\star$-$\lambda_{\mathcal{G}}$-closed, so by
Lemma 4.9, $A = (1,2)^\star$-$\mathcal{G}$-$Ker(A) \cap \tau_{1,2}$-$cl(A) = \tau_{1,2}$-$cl(A)$. Hence A is $(1,2)^\star$-α-closed.

Remark 4.14 The following examples show that concepts of $(1,2)^\star$-$\mathfrak{g}_{\alpha}$-closed sets and $(1,2)^\star$-$\lambda_{\mathcal{G}}$-closed sets are independent of each other.

Example 4.15 *In Example 4.12, we have $(1,2)^\star$-$\mathfrak{g}_{\alpha} C(X) = \{\Phi, \{b\}, \{c\}, \{b, c\}, X\}$ and $(1,2)^\star$-$\lambda_{\mathcal{G}} C(X) = P(X)$. In the space X, then the subset $\{a\}$ is $(1,2)^\star$-$\lambda_{\mathcal{G}}$-closed set but not $(1,2)^\star$-$\mathfrak{g}_{\alpha}$-closed.*

Example 4.16 *In Example 3.4, we have $(1,2)^\star$-$\mathfrak{g}_{\alpha} C(X) = \{\Phi, \{c\}, \{a, c\}, \{b, c\}, X\}$ and $(1,2)^\star$-$\lambda_{\mathcal{G}} C(X) = \{\Phi, \{a\}, \{b\}, \{c\}, \{a, b\}, X\}$. In the space X, then the subset $\{a\}$ is $(1,2)^\star$-$\lambda_{\mathcal{G}}$-closed set but not $(1,2)^\star$-$\mathfrak{g}_{\alpha}$-closed.*

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RESEARCH ARTICLE

An Efficient Framework for E-Learning System in Medical Education based on Internet of Things

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ABSTRACT

Learning management is the technique for valuating a student's knowledge or understanding of a particular topic of study. In this technological world, intelligent based learning system has become a more viable option to a range of people from beginners to get knowledge and the experts to get updated in learning. Recently, mobile based techniques have been applied within the learning methodologies for providing an effective learning environment. In this paper, a systematic review is carried out on application of mobile learning techniques to improve the medical education. Various techniques and its frameworks with respect to the E-learning, M-learning, techniques used, and the area of application in medical education are also discussed. This paper explores the applications of mobile based techniques for providing insights to the users and enabling them to plan, using the resources especially for the specific challenges in learning. Findings of this paper will guide the development of techniques using the mobile based technology as a source for handling learning methodologies more effectively.

Keywords: Medical education, M-Learning, Learning methodologies, Intelligent based systems

INTRODUCTION

The enhancement of student learning performance and satisfaction represents one of the main objectives of educational systems. Smart learning is a modern form in which smartphones, computers, and digital tablets are embraced by the learning system as aid for the method of teaching but not as a replacement for it [1, 2]. Smart learning is related to a modern type of education which relies on the application of technology, in virtual classes which can be participated from anywhere in the globe, and provides the material of the course with versatility and productivity at all times, because it is a constant, refreshed learning methodology that is accessible to the digital

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world [3]. While we advance with in field of AI, modern technologies are being framed for enhancing the efficacy of machine learning and make AI based applications such as deep learning and Artificial Neural Networks (ANN) [4] far-reaching and practical. To establish and sustain its view of the world, AI tries to minimize the human mind. It also includes algorithms that similar amount of user data-based results, allowing a computer to show behavior patterns learned from perceptions instead of social behavior. It allows mechanization to gain knowledge from information utilizing methodologies [5-10] and draw inferences and forecasts. Each new knowledge which is received by the AI-based system makes it much more convenient. E-learning can be defined as a digital learning program in which guidelines are created or prepared to assist learning and then sent to the targeted beneficiaries via electronic devices, which are typically computers or portable devices [11]. As illustrated in figure 2, e-learning can be constructed in two ways: one as a teacher based type of learning known as synchronous e-learning, and another as a personality based individual study known as asynchronous e-learning [11]. When students enroll in an asynchronous e-learning course which uses spoken or written texts in the form of drawings, pictures, visual effects, or multimedia as learning materials and includes assessments, the students are given the option to handle the time and location as well as the frequency at which they want to pursue their own learning [11], [12]. The other e-learning type, known as synchronous e-learning, is online and instructor based training where the instructions were to be presented or aided in real time by an instructor [11], [13]. This sort of e-learning method which is usually offered in online uses a variety of communication methods. Students that are completing the program typically connect on at a set time and communicate directly with the teachers [13]. Unfortunately, it seems that all these e-learning platforms such as asynchronous or synchronous have the same issues as traditional classrooms due to a lack of communication, which means that the diagnostic process among students and teachers cannot be correctly implemented. Furthermore, e-learning courses are available and developed for all students, regardless of their specific requirements and talents [14], [15].

Following are the provocations that are still under consideration from the perspective of intelligent based learning system:
1. Better understanding of the applications of soft computing based technologies in the learning system.
2. Effective implementation soft computing based techniques in the learning management. This should be done in order to identify the most effective methodology for learning.

The remainder of the paper is organized as follows. Section 2 depicts the definition of intelligent based learning methodology. Section 3 presents the applications of AI and E-Learning based online applications in Medical Education along with the summary of E-Learning based online applications in Medical Education. Section 4 presents the proposed model based on the soft computing for learning methodology. Section 5 depicts the limitations of this article. Conclusion and the future enhancements are shown in Section 6.

Intelligent Based learning
Intelligent based learning is an advanced form of education. It defines new paradigm for learning that perform the students to have an environment which is more effective for learning [25]. It also offers personalized technological environment for learning with the applications of computing based systems. Overall structure of intelligent based learning environment is shown in figure 2. This intelligent based learning offers the following features for the learners.
(i) It focuses on content of learners based on advanced computing based technologies
(ii) It is an intelligent based effective and tailored learning methodology based on advanced infrastructure of Information technology

Smart College in medical education
The smart college and smart class room is a form of initiative learning which facilitates the teachers to make the Information Technology (IT) as an integral part of the education. A smart classroom is a transformative area where the traditional ways of working can be converted to a digitalized way of working [26]. Activities of the learner should be connected with both the content of the subject and the applications of real world. The smart classroom

should also be categorized as a place with the combinations of various IoT based devices and its solutions that provide the way for learning analytics solutions, teaching which is learner-centered, and also to provide more personalized collaborative and creative learning strategies [27-30]. The strategy of smart classroom for E-Learning consists of co-dependent and interrelated components. Smart classroom makes the learners to adopt resources which are digitalized and it can communicate with the learning systems in any location and at any moment. It should also provide an active and necessary guidance for learning, tools for supporting or suggestions for learning in the in the right form, right place and at the right time.

E-Learning

Smart learning (S-learning) is an advanced form of education. It defines new paradigm for learning that perform the students to have an environment which is more effective for learning [4, 31-33]. It also offers personalized technological environment for learning with the applications of computing-based systems. Overall structure of smart learning environment is shown in **figure 1.**

This smart learning offers the following features for the learners.
(i) It focuses on content of learners based on advanced computing-based technologies
(ii) It is an intelligent based effective and tailored learning methodology based on advanced infrastructure of IT.

AI and E-Learning based online applications in Medical Education

M-Learning based learning consists of user perspective, verification, user interface, speech command-related applications and its recognition, controlling the attendance using an attendance application, gesture triggered commands etc. Communication in a smart classroom, among distinct devices and the consumer are treated by the AI based algorithms through the gateway [34]. A network gateway is placed between the devices depicts that the design of the AI based smart classrooms model is based on IP network such that every classroom must have a private IP network of its own for controlling the devices. Various works has been done by different researchers in the field of medical learning based on ANN. Authors in [35] proposed a customized multi-agent M-Learning framework based on the combination of Item Response Theory (IRT) and ANN. Adaptive tests and customized recommendations were shown in their system. These experts contribute to the learning situation with adaptation and intellect and serve as a human instructor who guides the learners through a well and personalized teaching situation. Authors in [36] introduced an ANN approach to identifying the learning styles of understudies [37]. Authors in [38] implemented the Updated DA model used to measure the output of student grades to acquire scoring from old datasets to determine new features using a NN.

Authors in [39] proposed a framework using the ANN, Multi-Layer Perceptron (MLP) and Radial Basis Function (RBF), by predicting the students attending an e-learning program with a success predictor. In their methodology, the students were tested online with a structured interview of 25 queries, administered with a query complexity as per the teaching principles. Each assessment was registered at the end in a unique database. Their method provides statistical information for each student during the online course duration relating to the evaluation operation **Table 1** highlights the current applications based on deep learning, technological advantages and drawbacks of each model in E-Learning.

Proposed model based on the AI based Smart Learning in medical education

User training data from multiple devices, AI and agent-based architectures, and smart learning-based technologies are all part of the suggested approach. The overall design of the proposed architecture is shown in Figure 2. The entire framework for evaluating different types of data gathered from multiple learning systems. This data is processed by combining AI and NN-based algorithms. The data management component and the content management tool were major parts of the AI-based system. The suggested system's main goal is to leverage AI-based technologies to process user inputs. It also serves as a platform for data analysis, summary management, and feedback management. By identifying user input using the free text technique, the AI-based report management module obtains, analyzes, performs, and triggers the action. All of the studied sources have been saved and changed

in the learning database. This module has a number of features, including learning-based information storage and a computational tool. The AI-based data analysis tool accepts input data, transforms it into information, and distributes it to the summary management module. The smart teaching method receives input on all of the requests and sources that were processed. Since the AI and learning approaches were merged, this model enhances the general accuracy of intelligent learning.

Implementation of the AI based Smart Classrooms for medical students
The spatial and technology design of an intelligent-based classroom are divided into two elements. The teaching facilities are evenly dispersed across the room, allowing the features of complicated permutations to be realized to the maximum extent possible. This can be accomplished in order to provide medical students with a dynamic learning environment. Some basic gadgets, such as the monitor, smart chalkboard, and floor, should be updated. The adoption of the smart classroom will prominence such as administration of learning resources, control of the educational environment, sharing of resources among medical students, and situation training.

Limitations

There were certain drawbacks to this AI-based smart teaching method, including: a huge section of the population, the system couldn't be too exact, the need to rely totally on technology, and numerous privacy and protection concerns. Exposure to a smart learning-based framework is denied to a substantial percentage of the population either to its size, their inability to acquire it, and the slowing competence and abilities to implement it. The technology cannot be too exact in order to exclude humans and their preconceptions. These applications have simplified the learning process, but they cannot take the place of tutors. Furthermore, no software, no matter how well-designed or technologically advanced, could ever be 100 percent accurate. These clever learning techniques frequently allow a customer to become entirely reliant on them. If the user loses their user id/password, all of the data may be lost for a short time or perhaps eternally. There's a chance that the privacy and security of the information stored in it could be compromised. Malicious users have the ability to alter data in such scenarios.

CONCLUSION

M-Learning is a computer based combination of software features and methods used in the process of delivering educational materials for medical students. This can be regarded as an integrative learning-based software system. Similarly, one of the most significant achievements for the intelligent-based learning system is the application of AI in learning. AI-based education systems can be utilized to enhance existing learning methods in the medical profession by providing personal services. This increases the learner's and faculty's skills by communicating with the teaching method as well as the medical student. This study provides a thorough examination of the many applications of AI-based learning approaches for medical students. All of the advantages of certain technical areas and components of learning approaches, such as smart classrooms, smart teaching, and smart learning, are detailed. The use of several AI models in the learning process, such as ANN, RNN, CNN, and Deep Learning based Neural Networks, is discussed. Several studies based on the application of AI in teaching and learning strategies have been conducted which including tailored learning models and self-learning. The role of various AI-based methods in the present M-learning management approach is also discussed.

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**Table 1**. Summary of E-Learning based online applications in Medical Education

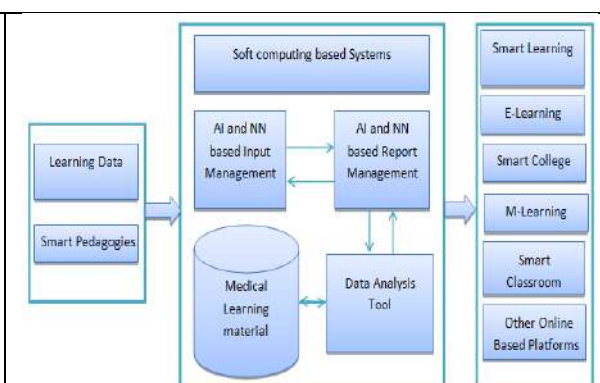| No	AI based Model	Main Area	Applications in Smart Learning
1	Virtual patient Learning	Online simulation platform that simulates an actual patient contact.	Students' communication and decision-making skills are assessed utilizing an online simulation platform that simulates an actual patient contact.[40]
2	Convolution Neural	Learning method based on Face	Expect the dependent variable of the tools for medical based learning. [36] [36]

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	Networks	Recognition	Usage of face expression to identify the thoughts of medical students [37]
3	Neural Networks	Speech recognition, feedbacks Prediction	Obtain the sequential events and interactions in the design Deliver a strong precision in recognition of speech & character and NLP based assignments [20]
4	Deep Learning based Neural Networks	Online presentations for medical students	Supports the virtual presentations more precisely.
5	Item response theory	Personalization of M-learning system	A customized learning system focused on multiple agents that provides the predictive tests and customized M-learning system [28, 31]
6	Trove Radiology Resident Dashboard	Automating the International Statistical Categorization of Diseases for Students better understanding	Provides a more up-to-date display for radiology students by automating the International Statistical Categorization of Diseases and Related Medical issues [41].
7	Deep belief Network	Natural language processing [33], speech recognition [32, 38]	This model Provide good accuracy in understanding the natural language and the speech recognition.
8	Artificial Neural Networks, K-Nearest Neighbor	Performance prediction of students	This model predicts the performance of students based on their scores in the M-learning management system [10, 11].
9	Self-Organizing Map and Back Propagation Neural Networks	Recognize the connection between both the content of learning and the learner's necessity for learning.	A combination of personalized AI based system which is used to select the learning objects for each medical student [9]
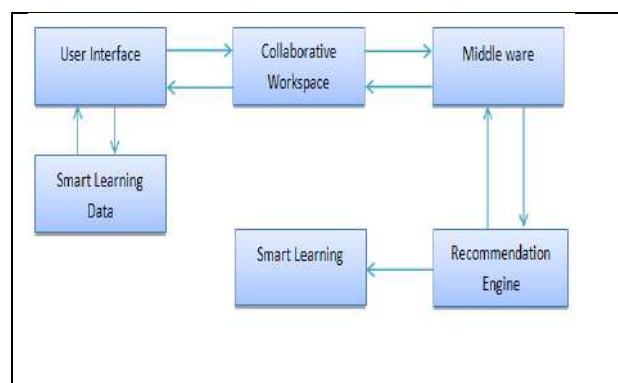
Fig.1. The overall structure of Mobile learning environment | **Fig 2. Architecture of the proposed AI based smart Learning system**

RESEARCH ARTICLE

Effects of Task Related Sitting Training on Balance and Functional Independence in Subacute Stroke Patients with Hemiplegia: A Randomized Sham Controlled Study

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ABSTRACT

Hemiplegic patients suffer from balance issues which lead to functional impairments after stroke. Several activities of daily living include sitting ability as a critical component. The study was aimed at evaluating the efficacy of a 4-week task related sitting training program in improving patients' balance and functional independence in such patients. 31 subacute stroke patients with hemiplegia were included in this randomized placebo-controlled study The subjects were included if they had first stroke within last 6 months and were able to sit. Subjects with orthopaedic, visual, cognitive-perceptual and other neurological deficits were excluded. The study group A (n=16) participated in a standardized seated reaching training program involving practice of task oriented reaching beyond arm's length along with the conventional therapy. The group B (n=15) received a sham training. Subjects were assessed before and after the completion of 4 weeks training using Total Berg's balance scale score (t-BBS), Functional reach distance (FRD) and Barthel index (BI). Both groups showed significant improvements in t-BBS and BI scores but FRD scores were not significantly improved in group B when within group comparison was done pre- and post- intervention. When between group comparisons were done, the participants from group A showed significant improvements in balance and functional independence scores at p<0.05 levels as compared to group B.The findings are suggestive of the efficacy of task related sitting training in improving the ability to balance during seated reaching activities as well as other activities of ADL which in turn results in improved functional independence.

Keywords: Stroke, Task related sitting training, Balance, Functional independence, Rehabilitation.

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INTRODUCTION

Poor balance and sitting ability are a common impairment after stroke [1-3]. Rehabilitation of balance function impairment after stroke is critical to independent living [4,5]. The normal control of balance is known to emerge as a result of integration of inputs from the vestibular, visual and somato-sensory systems; and aims to maintain the body's center of mass over the base of support [6]. Balance is believed to form essential foundation for all voluntary motor skills [7]. Balance disabilities have predictive value on comprehensive ADL function in stroke patients which means that early assessment and management of balance should be emphasized [8]. Available studies have mainly focused on balance impairments rather than balance disability and its impact on functional independence, i.e. static or dynamic balance while performing a tasks [9-11].Due to larger base of support and lower center of gravity, sitting provides more stability when compared to standing. Post-stroke muscle weakness and loss of dexterity leads to disability with poor sitting [2]. Ability to sit involves not only the ability to maintain the seated posture, but also the ability to reach, within and beyond arm's length, for different objects [2]. Individuals after stroke load their affected foot or activate the muscle of their affected leg insufficiently and are slower responding when they are reaching beyond arm's length in sitting position [11]. During reaching activities while sitting, muscle activation usually depends upon the amount of support which is provided to the feet. Whether both feet are supported on ground or not, trunk muscles are functionally active in stabilizing the upper body while it moves about over the base of support [12]. Furthermore, sitting ability has been shown to be one of the important prognostic indicators of outcome after stroke [3,8,13,14].

Intervention to train balance is a common focus of rehabilitation after stroke. Literature has demonstrated the efficacy of a sitting training protocol in individuals who had suffered stroke [2,12]. Previous work by Dean and Shepherd (1997) demonstrated effects of training for appropriate loading of affected foot on stroke patients and reported increased ability to reach further and faster. They demonstrated the efficacy of a sitting training protocol in individuals with post-stroke duration from 2 to 17 years [2]. Dean CM et al (2007) studied effects of sitting training on sitting ability, sitting quality and standing up early after stroke and concluded that the sitting training is both feasible and improves patients' condition in these aspects [12]. Cho G, Lee S & Woo Y (2004) reported improvements in task related circuit groups as compared to the conventional physical therapy group in functional task performance [15]. The research questions for this study were: (1) "Does completion of 4-week sitting training protocol improve balance ability associated with sitting?" (2) "Does completion of a 4-week sitting training protocol improve functional independence?"

METHODS

This randomized placebo-controlled study was conducted at the department of physical medicine and rehabilitation, Civil hospital, Ahmedabad, Gujarat. After ethical clearance from institutional ethics committee, subjects were recruited through convenient sampling. After screening for fulfilment of inclusion criteria and having the informed consent from all 31 subjects (M: 17, F: 14) and systematic randomization was done and the subject were assigned to the particular group according to their sequence of approach on 1:1 basis.Participants from group A performed a standardized training program which involved 10 daily practice sessions of reaching beyond arm's length for 30 minutes for 4 weeks in addition to the conventional physiotherapy [11]. The subjects used their unaffected hand for reaching, picking up and drink water from a glass from 3 reach direction conditions: (a) Forward, (b) 45o towards the unaffected side and (c) 45o across the body towards affected side. Subject practiced reaching while sitting on the height adjustable stool while feet were resting completely on floor. Height of the seat was adjusted to 100% of lower leg length. The glass was kept at height adjusted to 75% of shoulder height. The progression in training was done by increasing number of repetitions and complexity of task over 4 weeks' period. Each participant performed 250-350 reaches per session and average 3000 reaches over 4 weeks [11].

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Participant from group B received a sham training where cognitive-manipulative tasks were performed within arm's length for 30 minutes in addition to conventional physiotherapy. This sham intervention was added to avoid any effect due to placebo. Tasks were performed in completely supported sitting position with arm resting upon the table and workspace was confined to 50% of patient's arm length. The training was progressed over the 4-week period by increasing the number of repetitions and cognitive difficulty of cognitive-manipulative tasks. Thus, this training was unlikely to lead improvements in sitting balance and FRD [11].Participants of both groups participated in activities performed for approximately the same amount of time in the sitting position and performed an equivalent number of reaches. Both the groups were given conventional stroke rehabilitation including measures aimed at improvement of muscle force, range of motion, sensory function, flexibility and joint integrity; reducing tone and balance impairments and enhancing functional independence [16].Berg balance scale, functional reach test and Barthel index were used to assess improvements in balance and functional independence before and after the intervention. Assessor and patient blinding was implemented as therapist blinding was not possible. Wilcoxon signed rank test was used for within group comparison of outcomes pre- and post-intervention whereas Student's t-test was used for between group comparison of findings. Level of significance was kept at p<0.05 with 95% confidence interval.

RESULTS AND DISCUSSION

Both of the groups showed statistically significant improvement in t-BBS and BI total scores (p<0.01) when compared for within group, whereas FRD scores were not significantly improved in Group-B (p= 0.08) (table:1). When between group comparisons were done, the participants from group A showed significant improvements in balance and functional independence scores, expressed as t-BBS (p<0.03), FRD (p<0.01) and BI scores (p<0.05), compared to group B (table:2).Both the groups improved significantly in all three outcome measures but FRD showed statistically more significant improvements as compared to t-BBS and BI when between comparison of means was done. Previous studies by Dean CM et al (2007) [12]; Khallaf ME (2020) [17] and Bak and Lee (2021) [18] suggest that the sitting training protocol is both feasible and effective in improving sitting and standing up early after stroke and somewhat effective 6 months later. Many studies have proven efficacy of task related training in improving the ability to balance during seated reaching activities after stroke as well as improved sit to stand task along with less mediolateral sway when rising and sitting down.

Studies by Cheng et al (2001) [19]; Chen CI et al (2002) [20]; Rensink et al (2009) [21] and Kim and Lee (2013) [22] proved improvements in symmetry of weight bearing and distribution by task related training after stroke. Cho G, Lee S & Woo y (2004) proved improvements in symmetry of weight bearing distribution by task related training after stroke [15]. Salbach NM, Mayo NE, et al (2005) [23] has proved efficacy of task oriented walking interventions in improving balance self-efficacy during self-initiated gait activities. The results of the present study showing improvements in functional activities, sitting quality and functional reach performance by the sitting training along with the conventional therapy is in accordance with results of the studies mentioned above.

Studies by Dean CM et al (2007) concluded that individuals who were trained specifically to improve their sitting by focusing on appropriate loading of the affected foot were able to reach further and faster. They were able to increase the load taken through the affected foot and increased the consistency of activation of muscles in the affected leg. The carry over to standing up was observed [12]. Ahamad et al (2019) showed biomechanical similarities between reaching in sitting and the pre-extension phase of standing, which supports the carry over effects of seated reaching training to sit to stand and walking [14]. During sitting training, subjects practiced moving their trunk forward rapidly over the centre of mass whilst loading their legs. Although these components were practiced with the intention of improving sitting ability, they are also critical components of biomechanics of early phase of sit to stand activity. Khallaf ME (2020) reported that individuals who were trained specifically to improve their sitting by focusing on appropriate loading of the affected foot were able to reach further and faster. In addition, these individuals were able to increase the load taken through the affected foot and increased the consistency of activation

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of muscles in the affected leg [17]. However, it is not known whether this sitting training protocol is feasible and effective in improving trunk control and balance abilities associated with functions of daily living.

Present study supports the concept of specificity of training, which has been discussed in relation to the able-bodied subjects by Rutherford OM (1988) [24]and proposed as a means of rehabilitating the movement disabled by Carr and Shepherd [25]. The results of present study showing better improvements in the seated balance outcomes can be explained on the basis of the same mechanism as proposed by above mentioned case studies.Functional neuroimaging studies suggest that the functional gains produced in stroke patients by task related training are associated with increased activity in ipsileisonal primary sensory motor cortex and re-distribution of activity in several areas of sensorimotor network. This view is supported by the studies of Leipert et al (2000) [26], Nelles et al (2001) [27] and Jang et al (2003) [28]. Fujiwara et al (2001) used transcranial magnetic stimulation and suggested that recovery of trunk function following stroke is associated with increased activation of paretic trunk muscles by the unaffected hemisphere, suggesting role of compensatory activation of uncrossed pathways in recovery of trunk function [29].

The study has implications for rehabilitation, demonstrating that the stroke patients can improve their performance in functions of daily living by inclusion of short task related seated reach training that takes into account normative biomechanics related to trunk and lower limb function. It can be included in treatment intervention at an early stage of rehabilitation when there is greatest potential for neuroplasticity. As for all studies this study has its limitations, which include: (a). a smaller sample, (b). lack of long-term follow ups to confirm persistence of interventional gains, (c). exclusion of subjects who were not able to sit and reach, (d). lack of training for seated reach training on dynamic surface.

CONCLUSION

Task related sitting training can be used as an effective measure for improving sitting as well as functional balance during other activities when given with conventional treatment. Improvement in balance and functional abilities thereby leads to increased independence in activities of daily living. Therefore, it should be included early in treatment to gain maximum outcome benefits in short training period.

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Table 1: Pre- and post-intervention within group comparison of means

Group	t-BBS (mean + SD)			FRD (mean + SD) inches			BI (mean + SD)		
	Pre	Post	p-value	Pre	Post	p-value	Pre	Post	p-value
A (n=16)	33.31+ 7.55	44.37+ 6.11	<0.01	10.08+ 3.02	12.80+ 2.52	<0.01	59.42+ 12.82	78.12+ 11.76	<0.01
B (n=15)	34.80+ 5.33	44.20+ 5.68	<0.01	10.08+ 2.95	11.33+ 2.43	0.08	63.49+ 9.32	80.51+ 9.94	<0.01

Table 2: Pre- and post-intervention between group comparison of means of differences

Group	t-BBS			FRD (mean + SD) inches			BI (mean + SD)		
	Mean+SD	SE	p-value	Mean+SD	SE	p-value	Mean+SD	SE	p-value
A (n=15)	11.37+ 2.280	0.570	<0.03	2.72+ 1.079	0.269	<0.01	18.7+ 2.437	0.562	<0.05
B (n=16)	9.4+ 1.724	0.445		1.25+ 0.933	0.241		17.02+ 3.62	0.349	
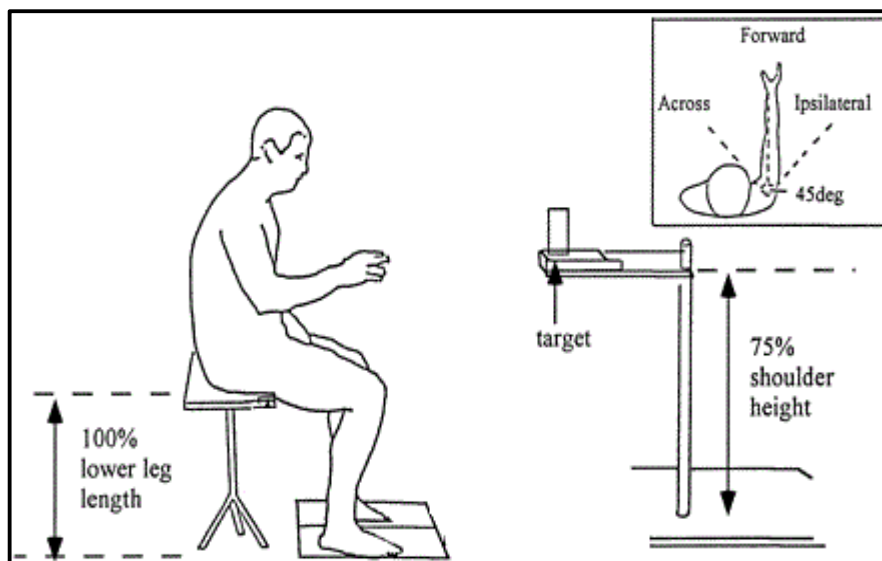
Figure 1: Schematic diagram showing seated reach out performance (Adapted from Dean & Shepherd, 1997)[2]

RESEARCH ARTICLE

Power Quality Impacts on a Grid-Connected Microgrid with Non-Linear Load and PV Integration

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ABSTRACT

In today's world, an increasing interest in renewable and alternative clean energy sources has been noticed. The self-renewing sources like solar, wind, hydro, etc. which can provide clean and ample amount of energy, can be used for electricity generation. The incorporation of these resources into the conventional power grid can supply clean and good-quality energy to the consumers. This can be done by integrating microgrids into our conventional power systems however, the power quality issues in a microgrid remain a major problem. The term Power Quality (PQ) can be defined as an occurrence that manifests as a non-standard voltage, current, or frequency resulting in the failure of customer equipment and hence, can have economic impacts on consumers. PQ issues arise in a microgrid when inconsistent natures of renewable energy sources are integrated with modern converter technology and also sometimes because of the existence of non-linear and unbalancing loads that are connected to it and these issues are becoming more and more significant with the development of highly sophisticated and sensitive devices. In this research work, an AC grid-tied microgrid model was developed in MATLAB/SIMULINK software to which various power quality events such as voltage sag, voltage swell, capacitance switching, transients, etc. are created, applied, and analyzed with the help of Fast Fourier Transform (FFT) and Wavelet Transform techniques. Also, the above-mentioned scheme was tested with a wide variety of system and fault parameters and the results so obtained are included in this paper.

Keywords: Microgrid, Renewable Energy, Power Quality, Power Electronic Converter, Wavelet Transform, Grid-tied Microgrid, MATLAB/SIMULINK.

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INTRODUCTION

A microgrid is a miniature and compact version of the conventional supply system that works either in low voltage or medium voltage networks and comprises a combination of micro-sources, energy storage systems, and various non-linear loads [1]. The energy storage systems consist of flywheels, super-capacitors, batteries, etc. that help in storing the excess energy produced by the microgrid system for use at a later time. The micro-sources constitute photovoltaic (PV) arrays, fuel cells, wind turbines, etc. which can be grouped to form units, each unit having only a small capacity which can then be integrated with power electronic devices and can be used at the utility sites [2]. In order to meet the large power demands of today's world with the conventional type generation systems, a tremendous amount of fossil fuels is being burnt on a daily basis which emits huge amounts of poisonous gases like methane, carbon dioxide, etc. into the atmosphere which in turn causes pollution and global warming phenomenon [3]. Also, since fossil fuels are non-renewable in nature, their natural stocks are depleting at an alarming rate. This is where the importance of renewable energy sources (RES) comes into the picture as their production is from natural sources found on earth such as solar, wind, hydro, and biomass [4]. Also, renewable energy sources are non-exhausting and replenishable in nature which makes them an excellent choice for power generation [5].

The term power quality can be defined as the net deviation of voltage or current waveforms of equipment from their pure sinusoid characteristics [6]. Power quality issues arise in a microgrid when inconsistent natures of renewable energy sources are integrated with modern converter technology and also sometimes because of the existence of non-linear and unbalancing loads that are connected to it[7]. Various power quality issues include voltage sags, swells, harmonics, and transients. The operation of a microgrid can be done either by connecting it to the main grid via the point of common coupling (PCC) called the grid-tied mode of operation or independently called the islanded mode of operation [8]. This paper mainly deals with the grid-tied operation mode of the microgrid. This research work aims at developing an IEEE-9 Bus System using MATLAB/SIMULINK platform and then modifying this system to accommodate the introduction of several faults at one specific bus of the system so as to analyze, in detail, the various effects these power quality events have on the operation of a grid-tied microgrid system.There are numerous works that have been done by researchers on the power quality issues arising due to the integration of renewable energy with a typical power grid, some of which are listed in Table-1 below.

Operational Process of the System

An IEEE 9-bus system along with a solar photovoltaic (PV) module was designed separately in the SIMULINK platform of MATLAB software. Then, a combination of PQ faults was introduced into the PV-integrated grid system by switching on two fault blocks at once, and then the simulation model was made to run by providing the desired running time. In the end, voltage and current waveforms were obtained for the faulted system for doing qualitative analysis. The voltage signals obtained from the above process were decomposed into two components i.e., approximate (high scale, low frequency) and detail (low scale, high frequency) signals by using Third Level Wavelet Decomposition Technique. Then, by applying Fourier Transform on the approximate signals, the magnitude and phase plots of the faulted system were obtained for doing quantitative analysis.

For the purpose of better understanding, the rest of the paper is arranged as follows. Section II gives a brief review of some of the works concerned with power quality issues in a microgrid that have been done by the researchers. Section III provides the basic idea behind PQ problems in a microgrid and explains these problems in a concise manner. Section IV gives a detailed operational process used to carry out this research work. Section V introduces the simulation modeling of different systems used in this work. Section VI analyzes the results of this experiment while section VII gives the conclusion and future scope of this paper.

Power Quality and Microgrid

A microgrid consists of a number of non-linear devices such as micro-sources, energy storage components, and converters which cause various power quality problems [18]. Power quality can be defined as the imperfections in

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the ideal sinusoidal characteristics of an equipment's voltage or current waveforms. The major power quality issues that are dealt with in this paper are transients, voltage sags and swells, harmonics, and unbalanced load switching. When sudden changes happen in a system due to any disturbance (may be intentionally or unintentionally) in its state, then it is called transient [19]. Transients take place within a very short time gap, ranging from some microseconds to 1 second. There are two types of transients i.e., impulsive and oscillatory transients that can occur in a microgrid. If the rms value of the supply voltage reduces to a range of 0.1-0.9 p.u. and remains like this for some time ranging between 0.5 cycles to a few seconds, then it is called voltage sag.When the rms value of the supply voltage rises to a range of 110-180 % and remains like this for a time ranging between 0.5 cycles to 1 min at power frequency, then it is called a voltage swell [20]. If there is a sinusoidal waveform of frequency 'w', then the different components of that waveform having frequencies 2w, 3w, 4w, etc. are known as harmonics. If all the different frequency components are added together, a quantity called total harmonic distortion (THD) is obtained [21].

Modeling and Simulation

For this paper, firstly a 9 Bus grid system was modeled to which various fault blocks were added to achieve the purpose of this paper. Then, to bus 2 of this system, a lumped model of the PV network was connected which acted as the DG for our microgrid model. The operation of the fault blocks was controlled with the help of Circuit Breakers (CB), whose activation and deactivation time was pre-set as per our desired introduction of various faults into the grid system. Then, a 400 kW PV System was designed separately in MATLAB/SIMULINK platform using four PV Arrays which were connected to a voltage source converter (VSC) via DC/DC converters and breakers. This in turn was connected to the main grid through a 400 kVA, 260 V/25 kV three-phase transformer. Finally, the voltage and current variations of bus 2 under different circumstances were observed and monitored so as to analyze the quality of power supplied by the microgrid model. The simulation diagrams of the PV system and the complete microgrid model are shown in fig. 2 and fig. 3 respectively.

RESULTS AND DISCUSSION

The System with Voltage Swell and Harmonics

In this case, both voltage swell and harmonics fault blocks were closed at an instant of 0.2 seconds i.e., 4000th sample onwards. From fig. 4, it can be observed that initially, the amplitude value in the magnitude plot increases to 0.9 but when the faults are introduced at 0.2 seconds, the amplitude value increases to 1.75 until the 8000th sample then further increases to attain the maximum value while the phase value in the phase plot initially decreased to -1.1 followed by a gradual increase for a short duration after which there were many sudden non-uniform variations.

The System with Capacitor Bank Switching and Harmonics

When both capacitor bank and harmonic switching fault blocks were closed at an instant of 0.2 seconds, it can be seen from fig. 5 that in the magnitude plot, the amplitude value increases initially to 0.9 and then becomes constant for 0.2 seconds. Then, the magnitude value decreases to 0.82 till the 8000th sample and further reduces to a constant value of 0.79. In the phase plot, the phase value initially decreases to -1.1, followed by a uniform and gradual rise to -0.8 value till 4000th sample. There are small disturbances at the 4000th sample after which again uniform rise occurs till the 8000th sample. Again, a series of disturbances are seen from the 8000th sample onwards.

The System with Voltage Sag and Harmonics

In this case, both voltage sag and harmonic fault blocks were closed at an instant of 0.2 seconds. It can be observed from fig. 6 that in the magnitude plot, the amplitude value increases initially to 0.9 followed by a drastic reduction to 0.45 value at the 4000th sample. Then, the value slightly increases to 0.49 and remains constant till 8000th sample, again decreases and finally becomes constant at 0.4 value while in the phase plot, the phase angle value initially decreases to -1.5, then increases gradually and uniformly till 4000th sample. We can see certain small spikes from 4000th till 5000th sample; then the phase value increases uniformly till 8000th sample. At the 8000th sample, again spikes are observed.

The System with Unbalanced Load Switching and Harmonics

When both unbalanced load switching and harmonic fault blocks were closed at an instant of 0.2 seconds, it can be seen from fig. 7 that in the magnitude plot, the amplitude value increases initially to 0.9 till 4000th sample and then reaches a disturbance period till 4500th sample. The value then decreases to 0.72, becomes constant till 8000th sample, and then further decreases to 0.62. In the phase plot, the phase value initially becomes -1, then increases gradually and uniformly till the 4000th sample. The phase angle value then becomes -0.8 at the 4000th sample. Further, the phase angle has uniform increment and then there are sudden disturbances at the 8000th sample.

The System with Three-Phase Fault and Voltage Swell

In this case, both voltage swell and fault blocks were closed at an instant of 0.2 seconds. It can be observed from fig. 8 that in the magnitude plot, initially the amplitude value increases to 0.9 then becomes constant till 4000th sample. After that, there is a series of increments and decrements in the value till the 8000th sample. Further, the amplitude value suddenly surges at 8000th sample and reaches its peak value at 9200th sample and then decreases while in the phase plot, the phase value initially decreases to -1.1 then increases uniformly till 4000th sample, then there is a series of increments and decrements in the phase value till 8000th sample. Further, the phase value suddenly surges at the 8000th sample, reaches its maximum value, and then decreases.

The System with Three-Phase Fault and Voltage Sag

In this case, both fault and voltage sag blocks were closed at an instant of 0.2 seconds. It can be seen from fig. 9 that in the magnitude plot, the amplitude value initially increases to 0.9, remains constant till 4000th sample, then decreases to 0.5 value and again becomes constant till 8000th sample. In the phase plot, the phase value initially decreases to -1.1 followed by a uniform increment till the 4000th sample. From the 4000th till 5000th sample, certain disturbances are seen. From the 5000th sample onwards, the phase value again rises uniformly to -0.5 till 8000th sample after which again disturbances are seen.

CONCLUSION AND FUTURE SCOPE

In this paper, a quantitative idea about the effects of different power quality events on a grid-connected microgrid was presented. From the quantitative analysis, a pinpointed idea was obtained on how exactly and at what point of time the PQ events and faults affected the system by studying the respective magnitude and phase plots. This research work may be extended by developing another MATLAB/SIMULINK model, considering other self-renewing sources like wind and hydropower and integrating them with solar PV so as to make a hybrid model. Power quality issues of the above model need to be studied and optimization may be done for better utilization when connecting to a microgrid.

ACKNOWLEDGMENTS

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Table-1: A Brief Review of the Related Literature

Reference Number	Issues Alleviated	Techniques Adopted
10	Unbalanced current and Harmonics	A four-leg VSI with a predictive type control method was used in the implementation of an active power filter that compensated the harmonics in addition to the unbalancing currents produced by single-phase non-linear loads.
11	Harmonics	PV grid-interfacing inverters connected to the distribution system that was regulated to act as harmonic-damping virtual impedance.
12	Harmonics	Passive LC low-pass filter interfaced with distributed generator (DG) via a three-phase inverter for mitigation of low order harmonics in inverter output voltage.
13	Harmonics	Harmonic filters connected in parallel with PV-integrated grid system to mitigate harmonics.
14	Transients	Grasshopper Optimization Algorithm (GOA) along with a droop control mechanism integrated to an islanded microgrid's PI control system to smoothen system transients.
15	Transients	Salp Swarm Optimization (SSO) algorithm incorporated with DG controller that achieved smooth transient response in a grid-tied microgrid.
16	Unbalanced loads	Modified Reinforcement Learning (RL) technique along with DSTATCOM that compensated the weakened AC supply due to the connection of unbalanced loads to a microgrid.
17	Voltage sags	Ensemble Empirical Mode Decomposition (EEMD) method that detected voltage sags in grid-tied and islanded microgrid modes.

Table-2: Magnitude and Phase Plot Variations with Different Fault Events

Combination of Faults	Magnitude Plot Variation			Phase Plot Variation		
	Initial	4000th Sample	8000th Sample	Initial	4000th Sample	8000th Sample
Voltage Swell + Harmonics	0.90	0.90	1.75	-1.1	-0.8	-0.65
Capacitor Switching + Harmonics	0.90	0.90	0.82	-1.1	-0.8	-0.39
Voltage Sag + Harmonics	0.90	0.45	0.49	-1.5	-0.8	-0.42
Unbalanced Load Switching + Harmonics	0.90	0.90	0.72	-1.0	-0.8	-0.30
Fault + Voltage Swell	0.90	0.90	1.50	-1.1	-0.8	-1.00
Fault + Voltage Sag	0.90	0.90	0.50	-1.1	-0.8	-0.50

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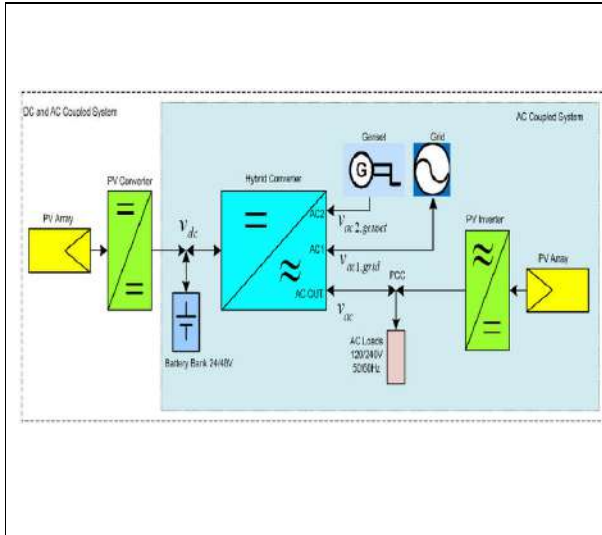
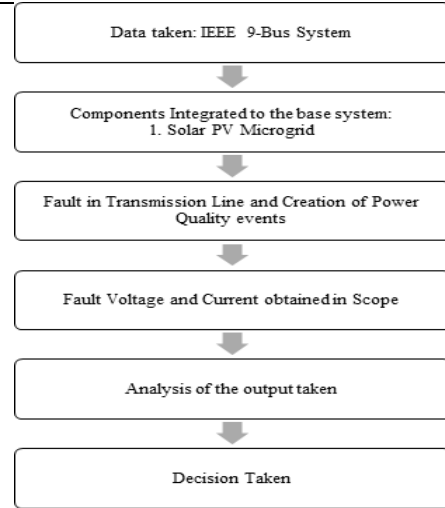**Fig. 1: Block Diagram of a PV-Integrated Grid System [9]**

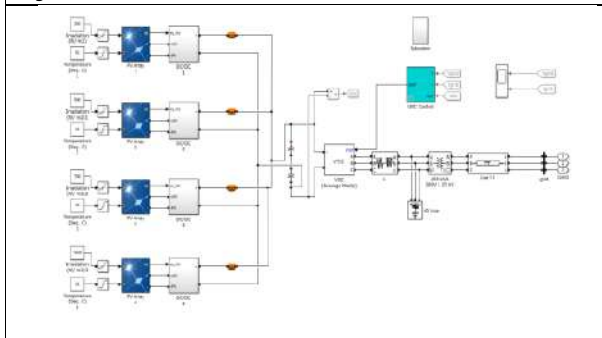**Fig. 2: Flowchart Depicting the Operational Process**

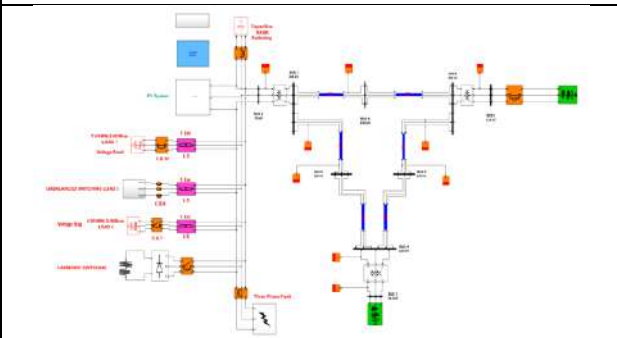**Fig. 3: Simulation Model of 400 kV PV System**

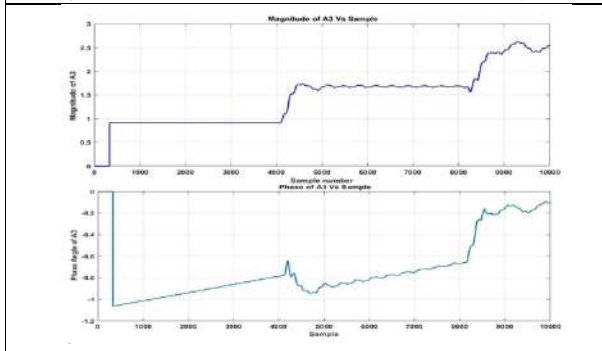**Fig. 4: The 9-Bus Microgrid Model**

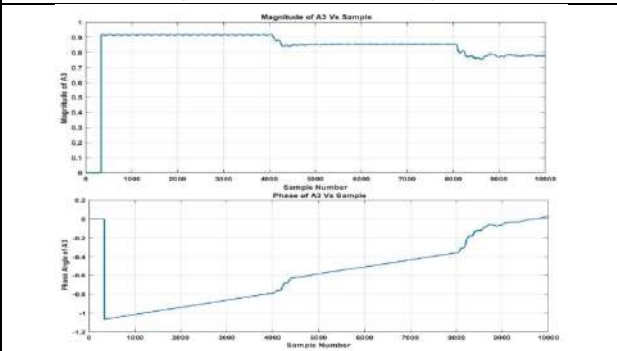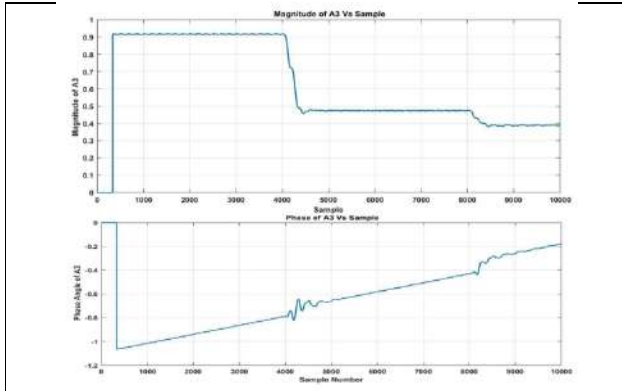**Fig. 5: Magnitude and Phase Plots with Voltage Swell and Harmonics**

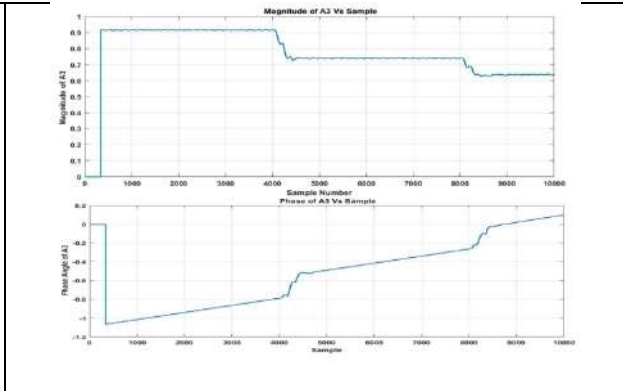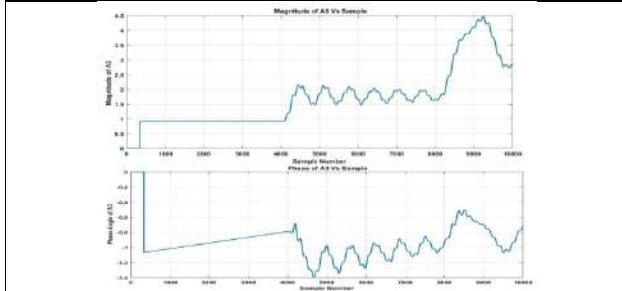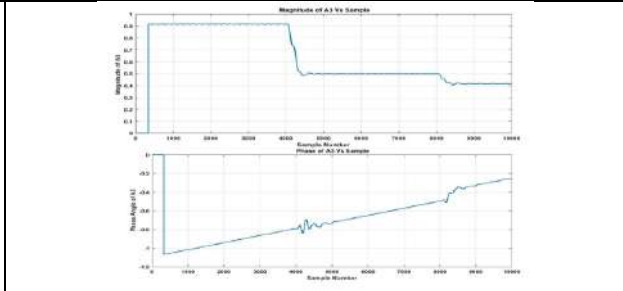**Fig. 6: Magnitude and Phase Plots with Capacitor Bank Switching and Harmonics**

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Fig. 7: Magnitude and Phase Plots with Voltage Sag and Harmonics

Fig. 8: Magnitude and Phase Plots with Unbalanced Load Switching and Harmonics

Fig. 9: Magnitude and Phase Plots with Fault and Voltage Swell

Fig. 10: Magnitude and Phase Plots with Fault and Voltage Sag

RESEARCH ARTICLE

Effect of Mixing Intensity for Biodiesel Production from Palm Fatty Acid Distillate

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ABSTRACT

Reaction parameters are the key indicators for the successful conversion of products. Mixing intensity is such parameter which can direct the conversion efficiency. In the present study, palm fatty acid distillate (PFAD) and methanol have been identified for studying the impact of mixing intensity w.r.t. biodiesel production. Novozyme 40013, an immobilized non-specific lipase from *Candida antarctica* is used as catalyst for the reaction. Mixing intensity was analyzed w.r.t change in concentration of free fatty acid, glycerides and biodiesel during the reaction. Reaction parameters used in the study were 60°C temperature, 5:1 molar ratio of MeOH: PFAD in the presence of 6% NS 40013 for 7 hrs by varying the mixing intensity from 300 to 800 rpm. 700 rpm has been identified as the optimum stirring rate for 93.2% biodiesel production. Study shows that mixing intensity has a great role for the biodiesel production from PFAD and methanol. Characteristics of biodiesel has been analyzed and showed good results.

Keywords: Mixing intensity, Palm fatty acid distillate, Biodiesel, *Candida antarctica*.

INTRODUCTION

Collision of reactants and catalyst play a significant role for the optimum production. Collision can be optimized by optimizing the mixing intensity between the reactants and catalyst. Variation of stirring rate or mixing intensity in the reaction system leads to the changes the concentration of reactants or intermediates or products. Mixing intensity

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w.r.t biodiesel production from various sources have been studied by several researchers. Peiter et al [1] studied the mixing intensity of soy biodiesel production and identified 350 rpm as the optimum level. Frascari et al [2] optimized mechanical agitation and evaluated the mass-transfer resistance in the oil transesterification reaction for biodiesel production. Brasio et al [3] studied the effect of mixing in biodiesel production through medeling and predicted the effect of mixing in the transesterification process. Stamenkovic et al [4] studied the effect of agitation intensity on alkali-catalyzedmethanolysis of sunflower oil. Present author also studied the effect of mass transfer kinetics for biodiesel production from jatrophacurc as oil using mathematical modelling [5]. Lakshmi et al [6] studied the mixing characteristics of the oil–methanol system in the production of biodiesel using different oils. The kinetic study and flow analysis of waste cooking oil was also studied by Janajreh et al [7] and showed that modelled results are similar with experimental results. Hossain et al [8] analysed the process parameters for biodiesel production where stirring was an important parameter. Biodiesel from castor oil was studied by Nazario et al [9] and identified the best conditions for a yield of 99.3%. Supramono et al [10] studied the effect of stirring for hydrogenation of oil and identified stirrer speed in between 650 and 800 rpm for optimum production. Tulliza et al [11] used static mixing reactor for palm oil biodiesel and showed that the mixing intensity can be enhanced by using number of modules where amount of catalyst can be reduced. Effect of mixing intensity for biodiesel production from different sources was also studied by Klofutar et al [12], Reyes et al [13], Sungwornpatansakul et al [14], Thompson and He [15]. But very few studies have been made for the effect of mixing intensity for enzymatic biodiesel production from industrial by product like palm fatty acid distillate. In the present research investigation, effect of stirring for biodiesel production has been analysed using palm fatty acid distillate and methanol in the presence of enzyme catalyst and identified the importance of mixing intensity during the reaction for optimum conversion of product.

MATERIALS AND METHODS

MATERIALS

PFAD was obtained from Emami Agrotech Ltd, Haldia, West Bengal. The enzymes used in the present study was Novozyme 40013, an immobilized non-specific lipase from Candida Antarctica with ester synthesis activity of 10000 propyl laurate unit/g. The chemicals monoglycerides and diglycerides were purchased from Scientific and Laboratory Instrument Co., Kolkata. Except otherwise specified all other chemicals were A.R. Grade.

METHODS

Initially 250 mL of crude PFAD was taken in an Erlenmeyer flask and heated up to 80^0 C to drive off moisture by continuous stirring for about 1 h. After that, mixing intensity was analysed through transesterification reaction with stepwise addition of alcohol in an appropriate proportion at a specified temperature for 8 hours maintaining other reaction conditions. Immobilized enzyme Novozyme 40013 was added as catalyst in definite proportion (w/w) in the reaction mixture. For minimization of deactivation of enzyme, stepwise addition of alcohol was allowed. For product analysis, definite amount of samples were withdrawn and centrifuged for 15 min to remove immobilized lipase. The supernatant part was taken in hexane and then evaporated to dryness and the products were isolated. The progress of reaction was observed by thin layer chromatographic (TLC) method after spotting the lipid mixture on a silica-gel G plate (0.2 mm thick) using hexane-diethyl ether-acetic acid (90:10:1) as a developing solvent. The lipid spots were identified by iodine absorption with triacylglycerol (TAG), diacylglycerol (DAG), monoacylglycerol (MAG) and biodiesel as standard. The yield of each reaction product after definite time interval with separate mixing intensity was determined separately by column chromatography using silicic acid as an adsorbent and 160 mL of hexane-diethyl ether: 99:1 as eluting solvent. After completion of reaction, the enzyme was washed with hexane, dried and reused for the next experiment. Biodiesel characterization was done according to the American Standard Testing Method (ASTM).

Values are reported as mean ± s.d., where n=3 (n=no of observation).

RESULTS AND DISCUSSIONS

Analysis of PFAD

The physicochemical characteristics of PFAD was shown in Table 1. It was observed from Table 1 that PFAD contains higher amount of FFAs which mainly includes palmitic acid and oleic acid. Among other acids, linoleic acid shares maximum amount alongwith stearic and myristic acids. Neutral glycerides namely triacylglycerols (TAG), diacylglycerols (DAG) and monoacylglycerols (MAG) are also present in PFAD which contributes $14.76\pm0.201\%$ in the composition. Little amount of unsaponifiable matters are also present in the PFAD which mainly includes sterols and tocopherols. Before enzymatic hydrolysis, PFAD was thoroughly bleached to remove peroxides.

Analysis of mixing intensity w.r.t. FFA concentration

Effect of mixing intensity w.r.t. FFA concentration of the reaction mixture directs the rate of conversion of product. Decrement of FFA in the reaction mixture enhances the increment of product percentage. Changes of mixing intensity from 400 to 800 rpm decreases the concentration of FFA due to the esterification reaction between FFA and methanol and transesterification reaction between neutral glycerides and methanol for converting methyl ester or biodiesel as observed in Figure 1. But beyond 700 rpm, there is no significant change in the FFA concentration as shown in Figure 1. This is due to the fact that enhance of mixing intensity does not always increase the collision between the reactants and the catalyst. So 700 rpm is the optimum mixing intensity for this transesterification reaction.

Analysis of mixing intensity w.r.t. glycerides concentration

Change of glycerides concentration w.r.t. mixing intensity for 7 hrs of reaction have been analysed maintaining other reaction conditions. It has been observed from Figure 2 that enhancing mixing intensity from 300 to 700 rpm changes the concentrations of TAG, DAG and MAG in the reaction mixture. Minor amounts of these components are also present in the final product. Enhancing rate of mixing intensity helps to disintegrate the component molecules and converts it to biodiesel. As observed form Figure 2, 700 rpm is the optimum stirring for the final product. In each mixing intensity, DAG content is highest compared to TAG and MAG till the reaction ends. It may be due to the fact that, during stirring, conversion of TAG to other intermediates or products occurs through DAG. So DAG content is high and mixing intensity has a specific role for the conversion of glycerides to the product. Table 2 shows the changes of total amount of glycerides during the reaction between. It has been observed from Table 2 that after 7 hrs of reaction there is a slight increase of glycerides concentration. This may be due to that fact that after 7 hrs of reaction, minor amount of biodiesel may be converted to glycerides as a reversible nature of the transesterification reaction.

Analysis of mixing intensity w.r.t. biodiesel concentration

Mixing intensity is also studied w.r.t. biodiesel conversion from esterification as well as transesterification reaction. It has been observed from Figure 3 that increasing mixing intensity enhances the conversion of biodiesel for 7 hrs of reaction and 700 rpm is the optimum stirring rate identified. Beyond that no further improvement of conversion has been observed. This may be due to the fact that higher stirring rate hampers the proper contact between reactants and active sites of enzyme which ultimately affects the production percentage.

Characteristics of biodiesel

Characterization of biodiesel from PFAD and methanol has been done after analyzing the mixing intensity and compared with the standards (biodiesel and diesel fuel) w.r.t. specific gravity, kinematic viscosity, density, acid value, calorific value, flash point, cloud point, etc., as shown in Table 3. It has been observed from Table 3 that the characteristics of biodiesel are quite comparable with biodiesel standards and diesel fuel in almost all the properties. Higher flash point of biodiesel compared to diesel fuel indicates that it can be handled and used safely than diesel fuel. So by optimizing the mixing intensity, good quality PFAD biodiesel can be prepared with standardization.

CONCLUSION

Mixing intensity between the reactants and catalyst plays an important role for the completion of reaction. Biodiesel production from palm fatty acid distillate and methanol depends on mixing rate for the successful conversion of reaction. Present study analysed the effect of mixing intensity w.r.t. free fatty acid, glycerides and biodiesel concentration in the reaction system. Analysis of the present study shows that proper stirring condition increases the productivity rate and is useful for the production of alternative energy sources maintaining identified parameters.

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Table 1: Physicochemical characteristics of PFAD

Component	Amount (% w/w)	Component	Amount (% w/w)
FFA (Total)	81.43±0.879	Palmitic acid	47.12±0.132
		Oleic acid	36.67±0.112
		Linoleic acid	9.21±0.043
		Stearic acid	4.76±0.019
		Myristic acid	2.08±0.054
Neutral glycerides	14.76±0.201	TAG	36.31±0.107
		DAG	42.51±0.118
		MAG	18.46±0.102
Unsaponifiable matters	3.7±0.012	Sterols	36.33±0.187
		Tocopherols	49.01±0.056
		Hydrocarbon and others	13.51±0.176

Table 2: Total glycerides during reaction

Time (hrs)	Total glycerides (% w/w)
0	14.76±0.201
1	11.65±0.042
3	8.43±0.011
5	4.21±0.039
7	2.37±0.024
9	2.53±0.016

Table 3: Characteristics of PFAD biodiesel

| Properties	PFAD Biodiesel	Biodiesel standard	Diesel fuel	Test method
Specific gravity (15 °C)	0.879±0.005	0.86 to 0.90	0.82- 0.95	ASTM D 6751-02
Kinematic viscosity (mm²/s) at 40 °C	4.61±0.015	1.96 to 6.0	1.3-4.1	ASTMD-445
Density at 15 °C (kg/m3)	876.5±0.162	865-900	820-860	ASTMD- 4052-96
Acid value (mg KOH/g)	0.39	0.8 max	----	ASTM –D 664
Calorific value (MJ/kg)	37.77±0.091	33 to 40	45	ASTM- 6751
Cloud point (°C)	1.53±0.009	5	---	ASTM D-2500
Flash point (°C)	194	>120	60-80	ASTMD-93
Cetane number	64.76±0.126	40 min	50	ASTMD-6751
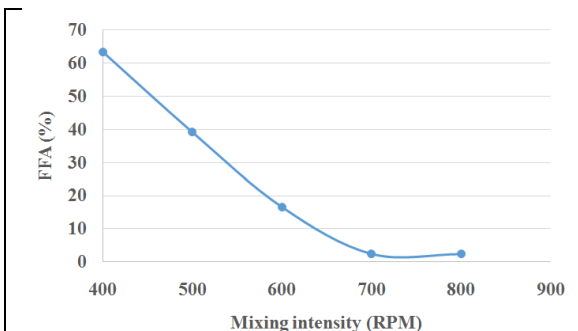
Figure 1: Effect of mixing intensity w.r.t FFA concentration. [Temperature: 60^{0}C, Time: 7 hrs, Molar ratio: 5:1 (MeOH: PFAD), NS40013: 6%]
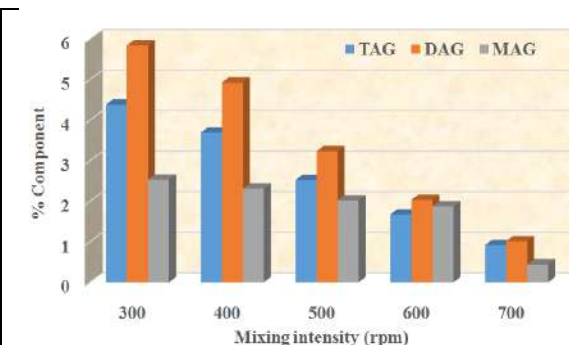
Figure 2: Effect of mixing intensity w.r.t glycerides. [Temperature: 60^{0}C, Time: 7 hrs, Molar ratio: 5:1 (MeOH: PFAD), NS40013: 6%]
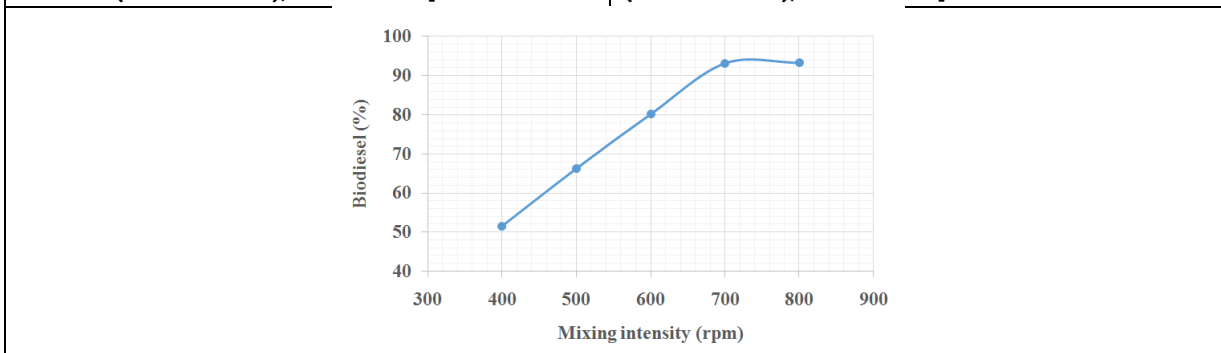
Figure 3: Effect of mixing intensity w.r.t biodiesel production [Temperature: 60^{0}C, Time: 7 hrs, Molar ratio: 5:1 (MeOH: PFAD), NS40013: 6%]

RESEARCH ARTICLE

Effect of Combination Pattern of Proprioceptive Neuromuscular Facilitation and Bowen Technique on Chronic Non Specific Low Back Pain, Functional Disability and Hamstring Flexibility in Housewives.

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ABSTRACT

Low back pain is one of the crucial health problems during life time with a prevalence of 80%, which causes functional loss and reduced productivity. There is a remarkable correlation between the low back pain and surrounding factors among housewife. Proprioceptive neuromuscular facilitation training is extensively used by physical therapists to treat chronic low back pain patients. Bowen technique is utilized to manage musculoskeletal conditions and works on the soft connective tissue of the body. 42 housewives (age between 30-60) were taken who have chronic non specific low back pain. They were divided into 2 groups. Group A(n=21) was given PNF, Bowen technique and Traditional physical therapy and Group B(n=21) was given Traditional physical therapy. Intervention was given for 5 days a week for 4 weeks. NPRS, ODI and VSR were taken before and after treatment. The result demonstrated that there was significant decrease in pain, functional disability and increase hamstring flexibility in both the group. But group A showed statistically more significant change in NPRS, ODI and VSR (p<0.05). This study concluded that combination pattern of PNF and Bowen technique is effective in decreasing chronic non specific low back pain , functional disability and increasing hamstring flexibility in housewives.

Keywords: Chronic non specific low back pain, Proprioceptive neuromuscular facilitation, Bowen technique, Oswestry disability index, Hamstring flexibility.

INTRODUCTION

Low back pain is most common musculoskeletal symptom. It was classified into 3 categories : acute , sub acute and chronic. Low back pain is developed by a disorder that affects the lumbar spine. It was described as pain located at the bottom of the twelfth rib and over the inferior gluteal fold with or without lower extremity pain. It was categorized as "specific" or "nonspecific" [1]. Low back pain is one of the crucial health problems during life time with a prevalence of 80%, which causes functional loss and reduced productivity [2]. In 85% of patients with low back pain, the signs and symptom are nonspecific without comprehensible diagnosis, prognosis, or treatment protocol [3]. Chronic low back pain is back pain lasting longer than 12 weeks. It has a negative impact on functional, socioeconomic, and occupational activities and on the psychological status [2].

Low back pain is not allocated to a distinguishable and recognized specific pathology like tumor, infection, inflammatory disease of spine, osteoporosis, radicular symptoms, fracture or structural deformity is called as Non specific low back pain. In the western world during the second half of the 20th century, it is one of the significant problems for public health systems and now it is extending worldwide [4,5]. In the general population around 70–80% of adults are accepted to experience minimum one episode of low back pain during their lives [6]. Previous studies shows that there is a remarkable correlation between the low back pain and surrounding factors among housewife [7,8]. The occupational workloads like bending, kneeling, body height and carrying heavy objects are associated with low back pain [9]. These activities expose the housewife to develop various occupational risks.Especially which are related to physical work overload as in the case of musculoskeletal impairment. Physical exertion caused by lifting or carrying heavy objects, repetitive trunk bending, trunk rotation and whole body vibration contribute to evolve low back pain [10]. Proprioceptive neuromuscular facilitation (PNF) training is extensively used by physical therapists to treat Chronic Low Back Pain patients. The PNF patterns are spiral and diagonal directions which are in obedience to topographic arrangement of the muscle being used in activities. PNF training has been recommended to facilitate muscle performance through its movement patterns [11].

PNF techniques can be divided in three categories: first, stretching/relaxation techniques, which includes hold-relax and contract-relax methods, second, the agonist muscle techniques, which include rhythmic initiation, combination of isotonics and third, the antagonist muscle techniques which include dynamic reversal, stabilizing reversal and rhythmic stabilization [12]. The Bowen technique is another type of physical manipulation entitled after Australian Thomas Bowen. It could be utilized to manage musculoskeletal conditions comprising acute sports injuries and works on the soft connective tissue of the body. It is soft and relaxing and does not utilize forceful manipulation. Bowen technique is executed on the superficial and deep fascia. The connective tissue that influences, envelops and detaches every tissues and organs in the body which is the part of the fascia [13]. The purpose of this study is to study the effect of combination pattern of proprioceptive neuromuscular facilitation and bowen technique on chronic non specific low back pain, functional disability and hamstring flexibility in housewives.

MATERIALS AND METHODS

An experimental study was conducted at sainath hospital, Ahmedabad. The sample size of the study consisted of 42 subjects with chronic non specific low back pain and they were divided into 2 groups of 21 in each group. Subjects were included in this study after taking demographic data, needed detailed physiotherapy assessment and subjects who fulfilled the inclusion criteria. Inclusion criteria of the study are (A) Age : 30-60 years, (B) Housewife, (C) Participants who Willingly participate, (D) Subjects who have non specific low back pain, (E) Subjects who have chronic low back pain (more than 3 months), (F) Housewife who do household activities at least for 3 hours/day, (G) Pain intensity 4 or >4 on NPRS scale. Exclusion criterias are (A) Subjects who have any neurological problem, (B) Subjects under fitness program, (C) Subjects suffering from severe medical illness, (D) History of any musculoskeletal disorders. (E) Pregnant women, (F) Radicular symptoms during functional evaluation.

Group A was treated with PNF, Bowen technique and Traditional physical therapy. Group B was treated with Traditional physical therapy. NPRS was taken before performing exercises. Score was recorded from the scale. The Oswestry disability index2.1a (Gujarati version) was given to the patient before performing exercises. Score was recorded from the questionnaire. V sit and reach test was performed before exercise protocol. Flexibility of hamstring muscle was measured. Intervention was given for 5 days a week for 4 weeks.

PNF was performed in three phases. Phase 1(week 1) was focused on rhythmic stabilization, phase 2 (week 2) was focused on combination of isotonics and phase 3 (week 3,4) was focused on chop and lift method. For Bowen technique, Patient was asked to wear loose shorts. This treatment was given on hamstring muscle. First at the side of the structure skin was pulled. Minimum pressure was applied to the outline of muscle to a point of resistance. This challenges the muscle and moves it out of its normal position. Distance between the thumbs and fingers is one inch so that the hands can move the muscles together where the hands were placed on the region. While sustaining gentle pressure in the medial site Following gentle rolling moves were done over the muscle. Each session was given for 20 minutes. This treatment was given thrice a week. Traditional physical therapy consist of Abdominal bracing, Abdominal bracing with heel slide, In quadruped arm lift with bracing, In quadruped leg lift with bracing, Diagonal curl up, Bridging, Isometric back exercise, SLR, Hip extensor exercise in prone, Back extensor exercise in prone, Hamstring stretching, Calf stretching. The subjects were asked to perform each exercise for 10 times with 10 seconds hold followed by 5 second rest between each exercise.

RESULT

This study involved 42 subjects aged between 30 to 60 years, who fulfills the inclusion criteria. These subjects were randomly divided into 2 groups and intervention was given in the form of Proprioceptive Neuromuscular Facilitation, Bowen Technique and Traditional Physical Therapy. The parametric test was used in statistical analysis because the distribution of data was normal. Paired t test is used to see the pre and post treatment effect. Unpaired t test was used compare both the groups. Table 1 shows mean age of both the groups. Table 2 shows paired t test of Group A and B pre and post mean of all outcome measures. Table 3 shows unpaired t test between Group A and Group B. Result showed significant improvement in all the outcome measures in Group A. The p value being < 0.05 showing extremely significant difference between the pre treatment and post treatment scores of NPRS, ODI, V Sit and Reach in group A.

DISCUSSION

The purpose of the present study was to see the effect of combination pattern of proprioceptive neuromuscular facilitation and bowen technique on patients with chronic non-specific low back pain , functional disability and hamstring flexibility in housewives. Result concluded that PNF, Bowen technique and Traditional physical therapy (Group A) significantly improves chronic non specific low back pain, functional disability and Hamstring flexibility in housewives based on the values of outcome measures. A study was conducted by ALTER in 1996, proprioceptive neuromuscular facilitation training comprises stretches and combinations of isotonics and the aim is agonist muscle facilitation so that it can produce the excitability of the motor neurons and increase the integration of additional motor neurons. The agonist muscle facilitation causes antagonist inhibition which assist to decrease in the excitability of the antagonist resulting in the muscular resistance in the facilitated muscle and the relaxation of the inhibited muscle. Previous studies has proven that range of motion increases by PNF training [14].

The hamstring muscles are related with low back pain and gait abnormality and also associated with motion dysfunction at the lumbar spine, pelvis and lower limbs. The study was done by Michelle Marr to see effects of Bowen Technique on hamstring flexibility over time stated that one session of bowen technique remarkably increased the flexibility of the hamstring muscle in asymptomatic individuals and also maintained this level of increase in hamstring flexibility for one week, showing continuing improvements. This study supports the current

study. There is increase in range of sit and reach test and increase in range of motion in phrase of popliteal angle after the implication of bowen technique for three alternate sessions, it showed that remarkable improvements in hamstring flexibility [13]. The present study showed that there is a significant decrease in NPRS score, ODI score and increase value in VSR score in Group B also in which intervention was given in the form of Traditional physical therapy. This is supported by the Systematic Review based on Exercise Therapy for Low Back Pain the study was done by Maurits van Tulder, which shows significant reduction of pain after conventional exercises in chronic low back pain [15].

CONCLUSION

This study evaluated the effect of combination pattern of proprioceptive neuromuscular facilitation and Bowen technique on chronic non specific low back pain, functional disability and Hamstring flexibility in housewives. Result concluded that PNF, Bowen technique and Traditional physical therapy (Group A) significantly improves chronic non specific low back pain, functional disability and Hamstring flexibility in housewives based on the values of outcome measures (NPRS, ODI, VSR) which was measured after given the interventions. Future of the study: 1.This study can be conducted to know the long term effects of these interventions. 2. The study can be carried out on subjects of different occupation groups. 3. This study only includes chronic non specific back pain individual so, other individuals can be studied.

Conflict of the study: Nil

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Table 1 shows mean age of both the groups.

AGE (FEMALE)	MEAN	SD
GROUP A	42.66	8.344
GROUP B	44.95	8.212

Table 2 shows paired t test analysis of group A and B.

	NPRS PRE MEAN±SD	NPRS POST MEAN±SD	ODI 2.1a PRE MEAN±SD	ODI 2.1a POST MEAN±SD	VSR PRE MEAN±SD	VSR POST MEAN±SD
GROUP A	5.52 ± 0.92	2.48 ± 0.75	40.61 ± 5.41	16.80 ± 6.43	18.21 ± 1.63	22.71 ± 1.75
GROUP B	5.14 ± 0.91	3.90 ± 0.94	38.14 ± 7.46	27.33 ± 7.14	18.38 ± 2.79	19.95 ± 3.07

Table 3 shows comparison of group A and B

	NPRS	ODI 2.1a	VSR
DIFFERENCE GROUP A	2.48 ± 0.75	16.80 ± 6.43	22.71 ± 1.57
DIFFERENCE GROUP B	4.0 ± 0.94	27.33 ± 7.14	19.95 ± 3.07

Graph 1 Graphical Presentation of Group A

Graph 2. Graphical Presentation of Group B

Graph 3 Shows Comparison of Both The Groups
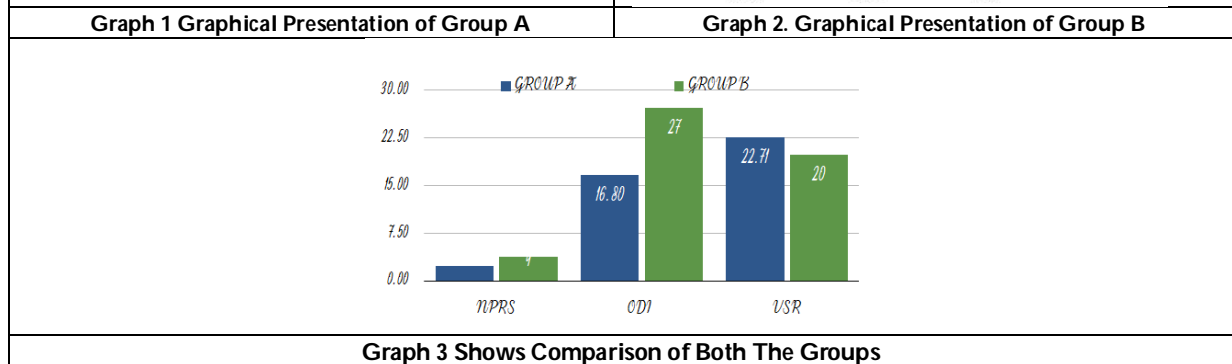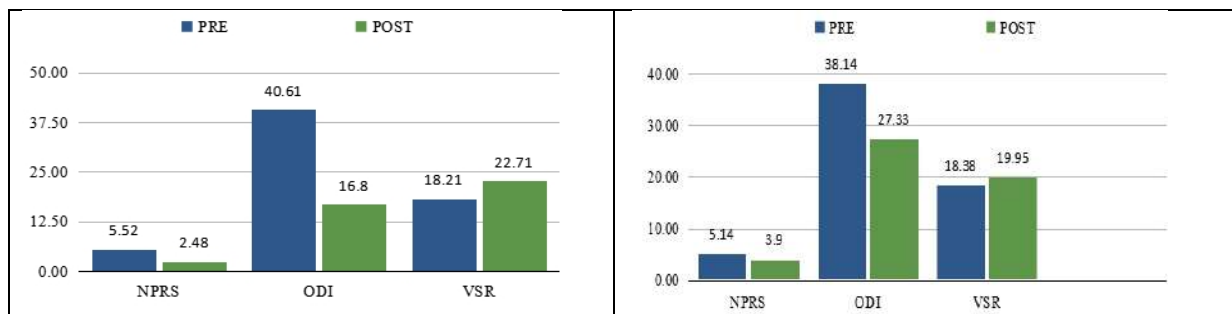
A Conceptual Framework for Ontology Learning from Text

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ABSTRACT

The semantic web is achieved by framing the précised data set which contains the knowledge of the domain and the common understanding of it. For arriving at a commonly understood domain knowledge, ontologies are developed based on the knowledge that is extracted. In the current scenario, there are different structures of data available all over the world and it is important to consider them while building ontologies. This paper proposes a framework to build ontology from unstructured data. The framework consists of three major phases with sub-phases for learning ontology from text. Natural Language Processing techniques are applied to pre-process the data and the component of ontology development is elaborated.

Keywords: Ontology, Ontology learning, learning techniques, NLP, Text Pre-processing

INTRODUCTION

The volume of information is increasing day by day as the number of internet users increases. The information is developed by different kinds of users from different sources. It is important to know the trustworthiness of the information and the real meaning of it. The semantic web technology helps to provide the meaning of data based on the knowledge. Semantic web is basically developed using ontologies. Ontologies play a vital role in framing semantic networks of data. Ontology is known for its knowledge of a specific domain that can be shared among different domains. The challenge here is to build ontology from unstructured data due to rapid increment of such data all over the world. The development of ontology requiring the intervention of humans and experts is not

possible all the time. The definition of ontology should be understood clearly before implementing it. The most-cited definition of ontology is, "a formal, explicit specification of a shared conceptualization" [1]. Ontologies define the concepts of the domain and relations between them. Different types of ontologies can be built viz., domain ontologies, task ontologies, application ontologies and top-level ontologies and so on. Building ontologies normally requires plenty of time, effort and human intervention. Ontology learning emerged to facilitate the task that helps the developers to build ontology semi-automatically or automatically.

Ontologies are built by using corpus, dataset, lexical, dictionaries and so on. In ontology, concepts are represented as classes and the verbs are represented as relations between the concepts. In ontology learning, the main role falls on keyword extraction which decides whether the ontology is right or not. It is very important to ensure the correctness of the key words to build correct ontology. Irrelevant keywords may lead to incorrect ontology. Normally, ontologies are developed by finding key words of a specific domain with their meaning. The collected terms and synonyms are arranged hierarchically and the relations are established. Finally, axioms are applied to produce knowledge-based content. The whole process is known as ontology learning. The processes are listed in ontology layer cake [2] as shown in figure 1. This research article proposed a framework to build ontology from unstructured text. The article is structured as follows: the state of the art of ontology learning is narrated in section 2. Section 3 describes the proposed framework followed by the case study in section 4. In Section 5 the conclusion is made and future enhancement of the research work is proposed.

Related Works
Ontology learning is the process of developing ontology from different sources like, structured data, semi-structured data and unstructured data. Usually ontology is built manually by the knowledge engineers with the help of the ontology development process [1]. To ease the task of ontology learning, it is important to learn the background and methods of building ontologies automatically or semi-automatically. Most of the existing research works are done by adopting the following major approaches viz: linguistic, statistical and logical [2]. Linguistic approach is implemented through pre-processing and relation extraction methods. Data mining, machine learning, and information retrieval are statistical approaches that can be used to extract domain-specific phrases, concepts, and correlations. Inductive Logic Programming (ILP) approaches, which include logic simplification and formal representation algorithms, are required for formal representation of a defined ontology [2].

Many surveys have been conducted on ontology learning [3][4][5][6] and they all conclude that to automate the ontology learning is a challenging task. The surveys depicted that there are existing ontology learning tools like Text2Onto [7], WebKB [8], DLLearner [9], HASTI [10]. Agnieszka Konys [11] made a commendable survey on ontology learning tools, which provides a detailed study on the knowledge repository of ontology learning. The researched article analyses 22 ontology learning tools under eight criteria. The work insisted that there is a need for the technology which enhances the dynamic development of ontology learning for fast adapting. Though there are many methods and tools for ontology learning, the present research work focuses on ontology learning frameworks. The frameworks for ontology learning from text are designed with hybrid approaches such as NLP, Statistical approach and Linguistic approach. The frameworks use the FCA and clustering techniques. The fuzzy and PSO algorithms were also used to extract concepts and concept hierarchies [12].

The ontology learning system frameworks can be compared with different dimensions. Different dimensions were used in the comparison of systems such as ASIUM, HASTI, Syndikate, SVETLAN, DODDLE II, Text-to-Onto and Web->KB, Ontolearn and OntoLT [13]. A domain-specific ontology was extracted from text using ontology learning tool Text2Onto. The tool applies an automatic extraction process using NLP techniques and learning algorithms. Initially, the tool extracted the 486 domain-specific concepts [14]. A framework BioOntoVerb was designed to prepare top level biomedical domain ontologies from the text. There are three different phases such as NLP Phase (POS Tagger, Syntactic Parser), Named Entity recognition phase (NE Recognition, Jape rules and gazetters) and Ontology Population phase (Semantic roles, Instance detection, Consistency checking, Instance classification) that

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were used to construct the biomedical domain ontology from the biomedical natural language texts [15]. An automated ontology generation framework contains five different modules such as Text processing approaches, Medical Semantic Annotation using N-Gram, Relation Extraction, Semantic Enrichment and Formal Ontology. This framework was linked with the biomedical domain of disease-drug domain to prepare the biomedical ontologies from text [16]. A framework was proposed to develop cognition ontologies from text by using SUMO (Suggested Upper Merged Ontology), WordNet and FrameNet [17].

The framework OntoLancs was suggested with four different phases to generate the ontology from text. The phases are, Domain Corpus (Lexical and Morphological Analysis and Semantic Tagging), British National Corpus (Basic concept List was prepared using Lexical and Text Analysis), Domain Taxonomy mapped with OWL and Ontology Edition [18]. The novel frameworks such as ADOL[19], OFIE[20], Word2Vec [21], and Evidential Reasoning Framework [22] focus on extracting text from the source converting them into PDF format and then the ontologies are built. The main objective of ontology learning from text is to minimize and ease the task of building ontologies from the text. The existing work seems to increase human intervention rather than reducing it. Existing studies revealed that there is a need for techniques to build ontology without the intervention of humans.

Conceptual Framework for Ontology Learning

In order to achieve the ontology learning from text, the related works show the essentials of defining a framework to build ontologies. As an outcome, a conceptual framework is proposed to build ontology from text. The proposed framework consists of two major phases; user interface and process with tools/technologies.

Phase 1: User interface: This phase allows the user to input the text to be built as an ontology. The resource may be any textual data of a specific domain.
Phase 2: Process: This phase actually involves the steps to build ontology with the help of tools and technologies. The phase contains four steps; information extraction, knowledge discovery, organization and evaluation.

Step 1: Information Extraction: Initially, the user or the knowledge engineer finds the appropriate corpus which has to be converted as an ontology. The corpus, which is the output of phase 1, will be taken as an input of phase 2 that will be passed to a pre-processing tool. The input resource can be taken from textual data such as textbooks, articles, web contents, structural data and search queries. The selected input sources is passed into the NLP assisted pre-processing tool and the keywords from the text are extracted. The NLP assisted pre-processed tool adapts the techniques such as Tokenization, Noise Removal, Stopword Removal, Stemming, Lemmatization and Normalization to identify and extract the seed terms from the inputted source. Tokenization involves the process of reification which breaks down the statements, phrase, paragraph or passage of a document into smaller units [23]. Each small unit is known as a token. The token can be formed in three ways; words, characters and subwords. Word tokenization is done by splitting the statement with the space as delimiter. For example, "Ready for every good work" can be classified into 5 tokens where each word is separated by space delimiter. Hence, the sentence will be tokenized as: T1 - ready, T2-for, T3-every, T4-good, T5-work. (T denotes token).

In character tokenization, each letter in the word is separated. For example, the word ontology can be tokenized like O-N-T-O-L-O-G-Y which has eight tokens. In the case of subword tokenization, the prefix or suffix of the word is identified and the token is framed. For example, the word higher can be tokenized as High-er which has two tokens. The noise removal is the process of removing special characters and symbols from the passage. The noise removal step is fully dependent on the domain. Example, Welcome!, the special character '!' will be removed and the output will be 'welcome'. The stop word removal helps to remove the conjunction word from the passage. Generally, articles and pronouns are classified as stop words which have no significance in the NLP tasks while extracting the terms. Stop words are frequently used in search engines, text classification software, topic modelling, and topic extraction, among other applications. For example, Original text: 'Ontology is a formal, explicit specification'. After removing the stop words, the above statement will be, ontology X where X is formal explicit specification. The

connecting words or the stop words are replaced with X, the dummy character. The concepts, relations and axioms are identified from the extracted keywords and the keywords are arranged hierarchically with the help of ontology learning algorithms.

Stemming is the process of extracting the root form of the verb by removing prefixes or suffixes. It reduces a word to its word stem, which affixes to suffixes and prefixes or to the lemma which is roots of words. Natural language understanding (NLU) and natural language processing (NLP) both benefit from stemming. Example: Writing - Wrote - Written can be stemmed to write which is the root term of all the above. Many algorithms are used to cut-down the prefixes or suffixes of the verb, where Porter's algorithm is the most common in the case of using English language contents. Lemmatization is the process of removing the inflections and mapping the word/text to the root text. It is similar to Stemming. The only difference is that lemmatization does the process in a proper way whereas stemming simply cut-down the verbs. For example, better can be mapped to good when applying lemmatization. When the stemming is applied, the output will be bet. Lemmatization uses a dictionary like WordNet for mapping the text or it applies some rule-based approach to find the appropriate root word of the text.

Finally, the Normalization technique is the process of transferring the text into canonical form. In this technique the appropriate word or the nearest word of the text is identified. For example, 'Gud', 'goood' can be transferred to 'good' which is the canonical form. In other ways, 'high-tech', 'high tech' can be framed as 'hightech' where the text is mapped to the nearest canonical form. The process of normalization depends on the tasks, since there is no proper way to do it. The common approaches used in text normalization are: spelling-correction based approach, statistical machine translation (SMT) and dictionary mapping. The other techniques are also used such as phrase structure and/or grammatical function parsing [24], semantic and discourse analyses [25], and part-of-speech tagging (POST) [26]. The NLP techniques are used to extract the keywords from the inputted text; the result is passed on to identify the knowledge discovery using ontology learning algorithms.

Step 2: Knowledge discovery is the process of extracting knowledge from data. It is achieved through ontology learning algorithms like lexical entry extraction, taxonomy extraction and non-taxonomic relation extraction to discover the concepts and relations between the concepts. The knowledge discovery process utilizes the output of the keyword extracted from the previous information extraction step as the input. Knowledge discovery can be done using the following process: i) terms/concept extraction, ii) relation extraction, iii) axioms and iv) evaluation.

Concept Extraction (CE): Concept extraction is a technique that identifies the most important text of the document. During concept learning, the information retrieval techniques are used to extract the words/phrases that only perform grammatical functions and words that are improbable to transmit domain-specific connotations are filtered. For example: Apple was founded by Steve Jobs. The text Apple and Steve Jobs are extracted as a concept.

Relation Extraction (RE): Relation extraction is a technique that identifies the semantic relationship between concepts. For example: Charles Babbage is the father of computers. The term 'is', 'the' and 'of' connect the terms Charles Babbage, father and computer which acts as a relation between concepts. The graphical representation of concept and relation is represented in the figure 3 where Cn and Cm refers to the concepts and Rx refers to the relations between the concepts.

Axiom: The extracted concept and relations are represented in a logical form that includes the rule. The logical form of ontology describes domain, range and properties of concepts with its relation. For example, the above-mentioned example can be defined as: domain: Cn, range: Cm, object Property: Rx.

Evaluation: Evaluation is the process that helps to assess the correctness of the ontology. The developed ontology can be evaluated using ontology evaluation tools. An ontology contains both taxonomic and factual information that need to be evaluated. The developed ontology is evaluated in the aspects of accuracy, adaptability, clarity, completeness, computational efficiency, conciseness, consistency and organizational fitness.

Step 3: Ontology Organization (OO): This process helps to increase the usability of the knowledge which is discovered by the ontology development process. The enormous amount of conceivable ontological classes and relationships are extracted from the learning process. Ontology organization seeks to achieve different goals such as, clustering synonymous terms and their relations, deriving inverse relations, discovering local centres of concepts and building higher-level ontologies. The ontology management tools such as Visuwords, Linked open vocabularies, WebVOWL, NavigOWL, Uni Lexicon vocabulary and Visual vocabulary Tematres are used in the organization of ontology.

Step 4: Ontology Evaluation (OE): Evaluation is the process of judgement/assessment of quality, value and importance. The Evaluation process also includes validation and verification processes. The prepared ontologies from text are evaluated using the tools such as Onto Analyser, Onto Generator, Onto Clean in WebODE, ONE-T and S-OntoEval.

CONCLUSION

Ontology learning is the process of building ontologies from structured data, semi-structured data and unstructured data. Since there is rapid generation of data from different sources by the different users, it is important to provide accurate content to the learner. Finding accurate content or providing valid content is a challenging task which requires huge human resources and more time to complete. The ontology learning from the text eases the task of providing accurate content based on the knowledge. The conceptual framework collaborating with NLP based pre-processing techniques is proposed in this article to build valid ontology from text. Supervised and unsupervised learning algorithms are used in ontology discovery and to extract the meaningful information from the text. In future, the proposed framework can be fully automated where the content of different sources can be automatically converted into knowledge-based resources. There is a wide opening in ontology learning from text research which is still at its development phase.

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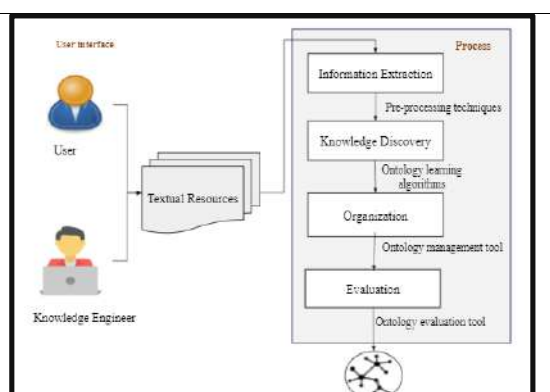
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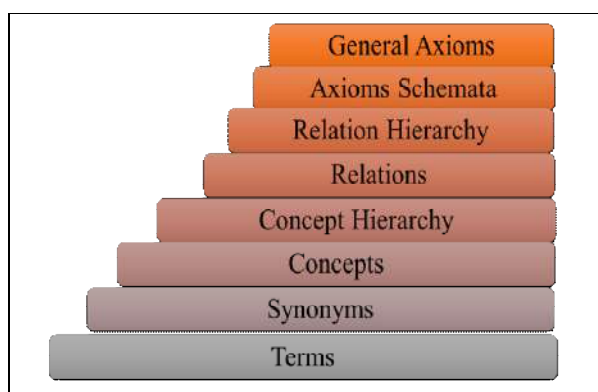
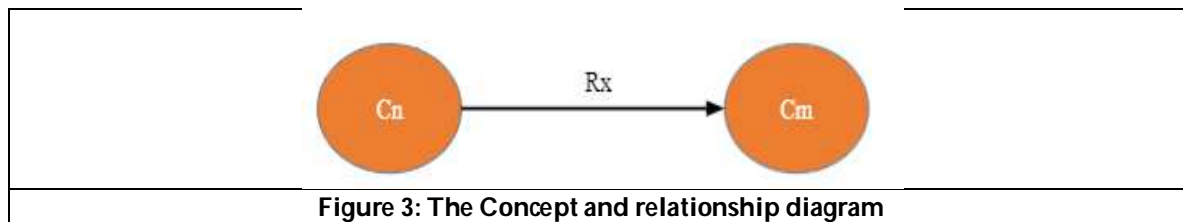| **Figure 1: Ontology learning layer cake** | **Figure 2: Conceptual Framework for Ontology Learning** |

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Figure 3: The Concept and relationship diagram

RESEARCH ARTICLE

Isolation and Screening of Pectinolytic Bacteria from Compost Soil

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ABSTRACT

Presently, industrial application of enzymes in biotechnological processes has expanded to a great extent. Pectinases have tremendous potential to offer to the industries especially food processing industries. Pectinolytic enzymes from microorganisms have gained lot of importance in recent times owing to their unique properties. Many microorganisms such as bacteria, yeast and moulds have been found to produce pectinase enzymes. In the present study, pectinolytic bacteria were isolated from the compost soil using pectin as the sole source of carbon and phosphorous. Five pectin degrading bacteria, showing distinct colony characteristics, were isolated and pure cultured. These bacteria were named as PDB01, PDB02, PDB03, PDB04 and PDB05. All the five bacteria were screened for pectinolytic activity by turbidity assay, plate assay, cup diffusion assay and turbidometric assay. Change in turbidity and zone of clearance observed in turbidity assay and plate assay respectively revealed the pectinolytic enzyme producing ability of these bacteria. The cup diffusion assay and turbidometric assay using culture filtrates showed the production of extracellular pectin degrading enzymes by these bacteria. The results of present study clearly indicated that the bacteria PDB01, PDB02, PDB03, PDB04 and PDB05 have significant ability to produce pectin degrading enzymes and could be employed for the production of pectinase enzymes but requires further more research.

Keywords: Bacteria, pectin degradation, pectinolytic activity, pectinase enzymes

INTRODUCTION

Enzymes are highly used in pharmaceuticals, biofuels, food and beverages, and consumer product industries. Microbial enzymes are enzymes produced by different microorganisms such as bacteria, yeast, fungi, etc. Among industrial enzymes obtained from microbes, 50% enzymes are obtained from fungi and yeast, 35% from bacteria and

15% from others [1]. The major characteristics of microbial enzymes are their capability and appreciable activity under abnormal conditions [2]. Some of the factors which make microbial enzymes strongly effective, attractive in many industrial processes include, its eco-friendly nature, reduced energy requirement and easy availability of raw materials for their production [3]. Pectin is a rigid polysaccharide found in the middle lamellae of plant cells and is broken down by pectinase enzyme. Pectinase is a general term for enzymes, such as pectolyase, pectozyme and polygalacturonase, commonly referred as pectic enzymes [4]. Pectinase enzyme constitutes 25% of the global food and industrial enzymes sales and the market is expanding day by day. Pectinase enzyme is required for the degradation of pectin. In fruit processing industry it helps in clarification of fruit juice [5]. Pectinolytic enzymes are used in the degumming of plant fibers, to remove pectic substances from pectic wastewater from vegetable and food processing industries and in tea fermentation [4]. The enzyme is also used to remove the mucilage from coffee beans [6].

The enzyme producing microorganisms are available in variety of sources such as soil, compost, agricultural waste, etc. Usually soil is a rich source of diverse microorganisms and many of them have the ability to produce various types of enzymes [7]. The compost soil (nutrient rich soil produced by microbial degradation of agricultural wastes) contains variety of microorganisms having the ability to degrade the polysaccharides such as pectin and cellulose. Pectin degrading microorganisms produce pectinase and such pectinase producing microorganisms are isolated from different sources such as compost soil, agricultural waste, orange peel, banana peel, etc., [8]. Isolation of pectinase producing microorganisms and production of pectinase from them has been regularly studied in recent years and production of pectinase has been reported from bacteria, fungi and yeast [9-13]. However, a thorough review of literature revealed that the search for better strains, especially bacteria, having the pectinase production ability is still required. In view of this the present research was undertaken with the following objectives - screening and isolation of pectinolytic bacteria from compost soil; study of pectinolytic activity of bacteria by assay methods; identification of bacteria based on biochemical tests.

MATERIALS AND METHODS

Isolation of bacteria from compost
Soil samples were collected from compost. The soil samples were transported to the laboratory in sterile polythene bags, air-dried and mixed thoroughly to make a composite sample. 1.0 g of soil sample was serially diluted and 0.1 ml of 10^{-1} to 10^{-4} dilutions were plated in duplicate on to 1% pectin agar plates and incubated at 37°C for 24-48 h [14]. After incubation the plates were observed for bacterial growth.

Study of pectinolytic activity of bacteria
Turbidity assay: The turbidity assay was carried out using 2% pectin broth tubes and pure culture of bacteria. 2% Pectin broth medium was prepared and 9.0 ml of broth was transferred to each tube, sterilised by autoclaving for 20 min. After sterilization the tubes were allowed to cool and aseptically inoculated with pure culture of all the five isolated pectin degrading bacteria, separately. One tube was maintained as control inoculated with 1.0 ml sterile distilled water. All tubes were incubated at 37°C for 24 h. After incubation, change in turbidity was observed by comparing with control tube.

Plate assay: Plate assay was carried out using 2% pectin agar plates and pure culture of bacteria. 2% pectin agar medium was prepared, sterilized by autoclaving and transferred to 90 mm Petri plates (20 ml / plate). After solidification, the plates containing pectin agar media were inoculated with five different bacteria, separately, by streaking and incubated at 37°C for 48 h. After incubation, iodine - potassium iodide solution (1.0 g iodine, 5.0 g potassium iodide in 330 ml water) was added to detect the clear zone [10, 15].

Cup diffusion method: The pectinolytic activity of bacteria was tested by using Kirby Bauer method [5, 12]. This is also known as cup diffusion method. 2% pectin agar plates and bacterial culture filtrate were used. 100 ml of 2% pectin broth medium was prepared in five different 250 ml conical flasks and sterilised by autoclaving. Each flask was inoculated with different bacteria and incubated at 37°C for 48 h. After incubation centrifugation was carried out

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at 6000 rpm for 10 min and culture filtrate was collected. Filtration of culture filtrate was carried out using Whatman No. 1 filter paper. Filter sterilized culture filtrates of all the five bacteria were used for further assay. Three wells were made at equal distance on each 2% pectin agar plate by using cork borer. After preparation of wells, 50 µl of culture filtrates of all the five bacteria were added to the wells in separate plates and to control well 50 µl of sterile distilled water was added and labelled accordingly. Assay plates were incubated for 24h at 30°C and after incubation flooded with iodine-potassium iodide reagent and these plates were kept undisturbed for 5-10 minutes. After 10 minutes plates were observed for clear zone around the well as an indication of pectin degradation.

Turbidometric assay: Turbidometric assay was carried out by using 2% pectin broth and bacterial culture filtrate [10]. 100 ml of 2% pectin broth was prepared. 8.0 ml of this pectin broth was transferred to 6 test tubes. 2.0 ml of culture filtrate of each organism was added in to five different tubes and labelled accordingly. To the control tube 2.0 ml of sterile distilled water was added. Optical density (OD) reading was taken for every one-hour interval at 450 nm for five hours. Graph was plotted by taking optical density on y axis and time on x axis.

Characterization of pectinolytic bacteria

Characterization of isolated pectinolytic bacteria was done by morphological studies and biochemical tests. The isolates were studied for morphological characteristics like colour, size, margin, form, elevation and texture. Gram's staining was done. All the bacteria were subjected for biochemical tests such as catalase test, indole test, methyl red test, Voges-Proskaeur test, citrate utilization test, urease test, gelatin and starch hydrolysis, and sugar fermentation test with glucose, sucrose, lactose and galactose [14].

RESULTS

Isolation of bacteria from compost

After 24 - 48 h of incubation, growth of bacterial colonies was observed on 1% pectin agar plates from compost soil sample (Figure 1). From this five different bacterial colonies that showed distinct colony characters were selected and named as PDB01, PDB02, PDB03, PDB04 and PDB05 (PDB – Pectin Degrading Bacteria). Selected bacterial colonies were pure cultured on to 1% pectin agar media plates and maintained at 4°C.

Study of pectinolytic activity of bacteria

Turbidity assay: After 24 h of incubation the result was observed in the form of change in turbidity. All isolated bacteria showed the positive result for turbidity assay by increased turbidity in pectin broth media due to bacterial growth (Table 1; Figure 2).

Plate assay: After 48 h of incubation good growth of all the bacteria (PDB01, PDB02, PDB03, PDB04 and PDB05) was observed on 2% pectin agar plates (Figure 3). The plates when flooded with iodine-potassium iodide solution showed clear zone around all the bacterial growth while no clear zone was observed in control plate (Figure 4). All the five isolated bacteria showed positive result for pectin degradation in plate assay.

Cup diffusion method: In cup diffusion assay, after incubation, clear zone was observed around the wells filled with culture filtrate of all the bacteria after treatment with iodine-potassium iodide solution indicating pectinolytic activity, while control well with sterile distilled water showed no zone (Figure 5).

Turbidometric assay: In turbidometeric assay, the OD values decreased with time interval (Figure 6) indicating the presence of pectin degrading enzymes in culture filtrate of all the pectinolytic bacteria screened.

Characterization of bacteria

Morphological characterization: The morphological (colony) characteristics like shape, margin, elevation, size, texture and colour of all the bacteria (PDB01, PDB02, PDB03, PDB04 and PDB05) are presented in Table 2. Gram staining revealed that all the isolated bacteria were Gram-positive rod-shaped bacteria.

Biochemical tests: All the bacteria gave positive result for catalase test by resulting in the production of effervescence on addition of H_2O_2. All the bacteria showed negative result for indole and Voges-Proskaeur tests. PDB02 and PDB04 were positive for methyl red and citrate utilization tests while PDB01, PDB03 and PDB05 were negative. All bacteria showed positive result for both gelatin and starch hydrolysis while negative result for urease test. In sugar fermentation tests all the five bacteria fermented glucose, sucrose, lactose and galactose by producing acid without gas formation (Table 3).

Based on the morphological and cultural characteristics (whitish, rough, dry, irregular, rhizoidal colonies; Gram positive rods) and the results of biochemical tests (catalase, gelatin hydrolysis and starch hydrolysis positive; indole test and urease test negative; fermentation of glucose, sucrose, lactose and galactose only with acid production and no gas production) the isolated bacteria were presumed to be *Bacillus* spp. However, the results of methyl red test, Voges-Proskaeur test, citrate utilization test were erroneous with respect to *Bacillus*. Therefore the precise identification of these bacteria requires further characterization such as molecular characterization by 16S rDNA analysis.

DISCUSSION

Microbial pectinase is considered as the most important enzyme for many industries. Varieties of microorganisms with the ability to produce both extracellular and intracellular enzymes have been isolated and identified. Some of the factors which make microbial enzyme strongly effective, attractive in many industrial processes include, its eco-friendly nature, reduced energy requirement and easy availability of raw materials for their production [3]. Pectinase enzyme is produced by both prokaryotic and eukaryotic microorganisms. Bacteria have the ability to produce extracellular pectinase enzymes and these enzymes are essential for the decaying of plant material. It has wide range of applications in the field of biotechnological industries, fermentation and wastewater treatment [6]. In the present study, pectin degrading bacteria (PDB) were isolated from compost soil using pectin agar media. Five different morphologically distinct colonies were selected and were screened for pectin degrading ability by turbidity assay, plate assay, cup diffusion assay and turbidometric assay. All the five isolates showed good growth on pectin agar and pectin broth media. All the bacteria studied produced the clear zone around the growth on treatment with iodine - potassium iodide solution indicating the ability to produce pectinase enzyme and thereby pectinolytic activity. The culture filtrates of all bacteria also exhibited pectin degrading ability showing the capability of bacteria to produce extracellular pectinolytic enzymes. Comparatively bacteria PDB02, PDB04, and PDB05 showed more pectinolytic activity than PDB01 and PDB03 isolates.

Soil especially in and around compost are good sources of biodegrading bacteria including pectin degrading bacteria. Pectinolytic bacteria were isolated and screened from agricultural waste dump soils in Vellore, Tamilnadu, India [3]. The isolation of pectinolytic bacteria was carried out using soil sample and rotten orange fruit, and screened for pectinolytic activity [12]. Pectinolytic bacterial isolates were isolated from Marand city farmlands, Iran and screened for pectinase activity [13]. Pectinolytic bacteria were isolated from decomposing fruit materials and pectinolytic activity was screened [16]. Pectinolytic *Bacillus subtilis* SAV-21 was isolated from fruit and vegetable market waste soil and production of pectinase by solid state fermentation was studied [17]. 1% and 2% pectin agar and broth media were used in the present study. One percent pectin concentration showed the maximum pectate lyase and pectin esterase activity by *Bacillus* sp. [18]. Plate assay, turbidity assay, cup diffusion assay and turbidometric assay are regularly used for screening of the pectinolytic activity of microorganisms including bacteria [5, 10 - 12, 15].

The isolated strains were Gram positive bacillus and based on morphological characterization - the bacterial colonies appeared in white colour with undulate filamentous entire and rhizoid margin with irregular shape. All five bacteria showed positive result for catalase, starch, and gelatin hydrolysis; all five isolates had the ability to ferment glucose, galactose, sucrose and lactose by producing acid without gas production. Two isolates showed the positive result for

citrate utilization and methyl red and all bacteria showed negative result for VP test, urease and indole. Based on morphological and biochemical tests the strains could not be precisely identified but presumed to belong to genera *Bacillus*. Molecular characterization using 16S rDNA analysis could be helpful in precise identification of the isolated bacterial isolates. Many researchers have employed morphological and biochemical tests for the identification of bacteria. Based on staining and biochemical tests pectin degrading bacteria were identified as *Bacillus* sp. [5]. Based on the results of morphological and biochemical tests, the isolated pectinolytic bacteria was identified as *Bacillus* sp. [9, 19]. Molecular methods have been employed for precise identification of pectinolytic bacteria up to species level. Based on molecular studies three bacteria were identified such as *Enterobacter* MF41, *Enterobacter* MF84, and *Enterobacter* MF 90, which produced pectinase enzyme [13]. Pectinase producing *Bacillus subtilis* SAV-21 [17] and *Bacillus* sp.MFW7 [20] were identified by 16S rDNA sequencing.

CONCLUSION

In the present investigations, bacteria were isolated on pectin agar media and screened for their ability to degrade pectin. Five strains recorded significant pectinolytic activity. These strains were further subjected for identification using morphological and biochemical tests. The result of all these tests revealed that the isolated bacteria might belong to *Bacillus* genera. However, precise identification requires further molecular studies such as 16S rDNA analysis. The present study has been successful in isolating pectinolytic bacterial strains and in their preliminary screening and identifying their ability to synthesize pectinolytic enzymes. Better utilization of these isolates requires further research in characterization of them; isolation, purification and characterization of enzymes, etc.

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Table 1: Turbidity assay of pectinolytic bacteria

Bacteria	PDB01	PDB02	PDB03	PDB04	PDB05
Turbidity	+ + +	+ +	+	+ + +	+ + +

Table 2: Colony characteristics of pectinolytic bacteria

Bacteria	Shape	Margin	Elevation	Size	Texture	Colour
PDB01	Filamentous	Filamentous	Flat	Moderate	Dry	White
PDB02	Irregular	Undulate	Flat	Moderate	Dry	White
PDB03	Circular	Entire	Flat	Large	Dry	White
PDB04	Rhizoid	Rhizoid	Flat	Moderate	Rough	White
PDB05	Rhizoid	Rhizoid	Flat	Large	Rough	White

Table 3: Characterization of pectinolytic bacteria by biochemical tests

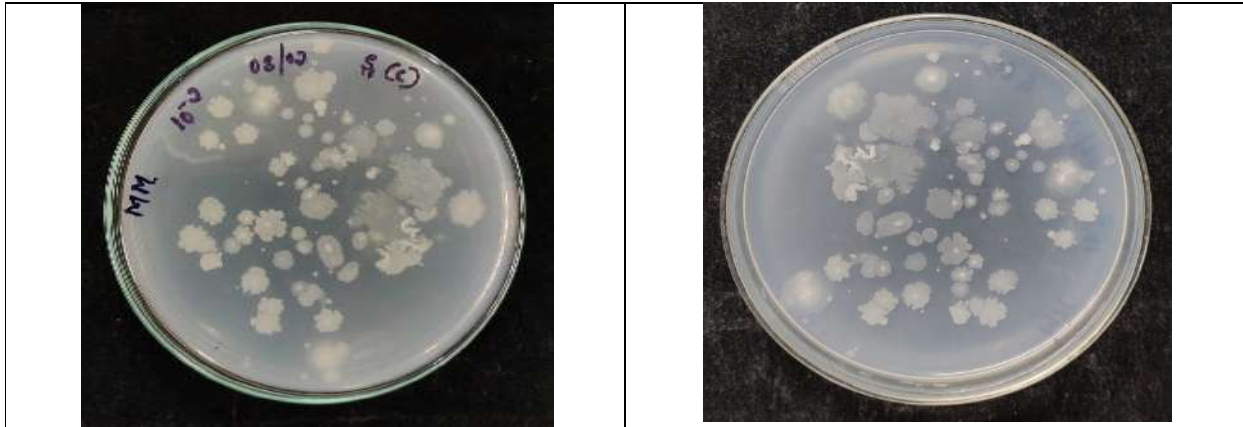| Biochemical tests	PDB01		PDB02		PDB03		PDB04		PDB05	
Catalase	+		+		+		+		+	
Indole	-		-		-		-		-	
Methyl red	-		+		-		+		-	
Voges- Proskauer	-		-		-		-		-	
Citrate utilization	-		+		-		+		-	
Gelatin hydrolysis	+		+		+		+		+	
Urease	-		-		-		-		-	
Starch hydrolysis	+		+		+		+		+	
Sugar fermentation	Acid	Gas	Acid	Gas	Acid	Gas	Acid	Gas	Acid	Gas
Glucose	+	-	+	-	+	-	+	-	+	-
Sucrose	+	-	+	-	+	-	+	-	+	-
Lactose	+	-	+	-	+	-	+	-	+	-
Galactose	+	-	+	-	+	-	+	-	+	-

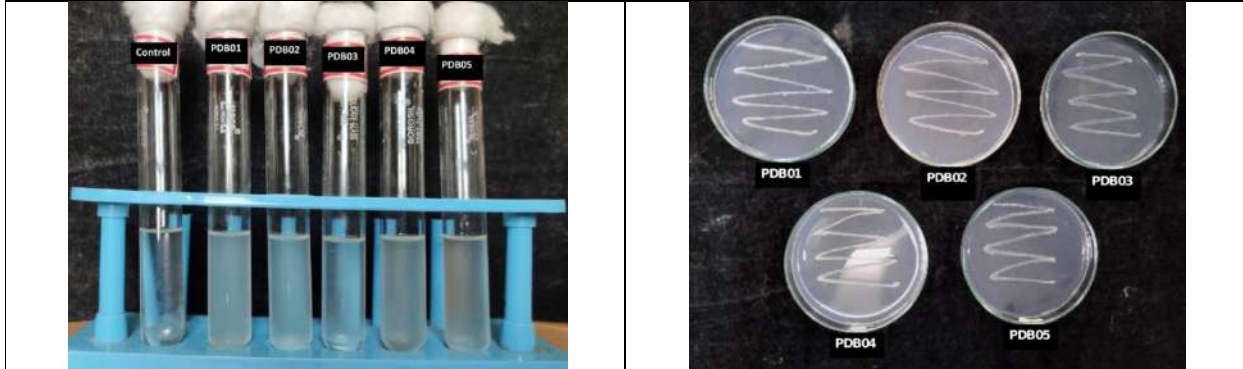**Figure 1: Bacterial colonies from compost soil on 1% pectin agar plates**

Figure 2: Increased turbidity indicating the growth of pectinolytic bacteria in 2% pectin broth

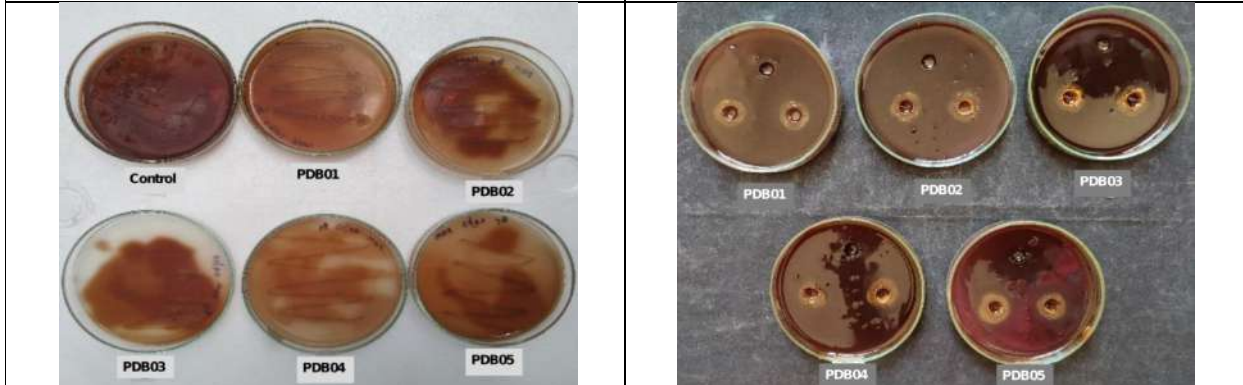**Figure 3: Growth of pectinolytic bacteria on 2% pectin agar plates**

Figure 4: Clear zone around the growth of pectinolytic bacteria on 2% pectin agar plates on treatment with iodine-potassium iodide reagent

Figure 5: Cup Diffusion Assay - clear zones around the wells (bottom) having culture filtrates of pectinolytic bacteria on 2% pectin agar plates on treatment with iodine-potassium iodide reagent; Top wells – Control
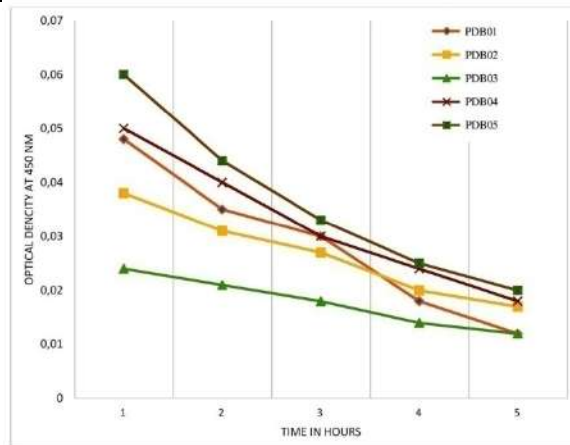
Figure 6: Turbidometric Assay – Graph showing the decrease in turbidity of pectin broth inoculated with culture filtrates of pectinolytic bacteria with time

RESEARCH ARTICLE

Cobalt(II), Nickel(II) and Copper(II) complexes of Heterocyclic Based Ligand: Synthesis, Spectral Characterization, Antimicrobial, Antioxidant And DNA-Binding Studies

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ABSTRACT

A new series of transition metal complexes of type [ML_2X_2], where M is Co(II), Ni(II) and Cu(II) L is 4-methylaminopyridine and X is thiocyanate ion were synthesized and characterized by various spectral and physico-analytical techniques such as elemental analysis, magnetic, UV-visible, FT-IR , EPR and TGA. The studies envisaged a hexa-coordinated geometry for the complexes, where the 4-methylaminopyridine and cyanate ion OCN^- ligand act as a monodentate manner *via* nitrogen atoms of 4-methylaminopyridine heterocyclic and cyanate ligands. The free radical scavenging activities of the complex and the ligand have been determined by measuring their interaction with the stable free radical DPPH. The complexes have larger antioxidant activity as compared to the ligands. DNA-binding properties have been studied by fluorescence-emissions   method. The outcome suggests that the metal complexes strongly bind to DNA because of metal complexes are well-known to speed up the drug action and the ability of a therapeutic agent which can frequently be enhanced upon coordination with a metal ion.

Keywords: 4-Methylaminopyridine, OCN^- ion, antioxidant and DNA-binding.

INTRODUCTION

Heterocyclic compounds are the cyclic organic compounds which contain at least one hetero atom, the most common heteroatom's are the nitrogen, oxygen and sulphur but heterocyclic rings containing other hetero atoms are also widely known. Heterocyclic compounds considered one of the vital classes of organic compounds, which are used in many biological fields, due to it is activity in multiple illnesses. Biological molecules such as DNA and RNA, chlorophyll, hemoglobin, vitamins and many more contains the heterocyclic ring in major skeleton. There are a lot of heterocyclic compounds which are have application in many common diseases such as; triazine derivatives have been used as antimicrobial herbicides, urinary antiseptics and anti-inflammatory agents. Benzimidazole derivatives have been reports to possess wide range of biological activities such as antibacterial, antifungal, antiviral and anthelmintic, etc. [1,2].

Heterocycles have been found a key structural in medical chemistry and also they are frequently found in large percent in biomolecules such as enzyme, vitamins, natural products and biological active compounds including antifungal, anti-inflammatory, antibacterial, antioxidant, anticonvulsant, antiallergic, enzyme inhibitors, herbicidal activity, anti-HIV, antidiabetic, anticancer activity, insecticidal agents. 4-methylaminopyridine is heterocyclic compound. The present study aims at the microwave assisted synthesis and spectral characterization of cobalt(II), nickel(II) and copper(II) complexes of heterocyclic based ligands.

EXPERIMENTAL METHOD

Materials
Cobalt nitrate, nickel nitrate, copper nitrate, 4-methylaminopyridine and sodium cyanate were purchased from Alfa Aaser Company and used as such. The organic solvents used, viz., DMSO, DMF, CH_3OH and CH_3CH_2OH were of AnalaR grade and used as such without further purification.

Synthesis of Co(II) complex
1.52 g (13.82 mmol) of 4-methylaminopyridine in CH_3OH and 4.50g (6.90 mmol) of sodium cyanate in C_2H_5OH were added to the methanolic solution of cobalt nitrate 1.00g (3.40 mmol) and this was followed by microwave irradiation for few minutes after each addition by using IFB 25 BG-1S model microwave oven. The resulting precipitate was filtered off, washed with 1:1 C_2H_5OH: CH_3OH mixture and dried under vacuum. A pink colored complex was obtained with the yield of 35.75%.

Synthesis of Ni(II) complex
1.52 g (13.82 mmol) of 4-Methylaminopyridine in CH_3OH and 4.50g (6.90 mmol) of sodium cyanate in C_2H_5OH were added to the methanolic solution of $Ni(NO_3)_2.6H_2O$ 1.00g (3.40 mmol) and this was followed by microwave irradiation for few minutes after each addition by using IFB 25 BG-1S model microwave oven. The resulting precipitate was filtered off, washed with 1:1 C_2H_5OH: CH_3OH mixture and dried under vacuum. A green colored complex was obtained with the yield of 24.71%.

Synthesis of Co(II) complex
0.71 g (6.45 mmol) of 4-methylaminopyridine in CH_3OH and 0.54g (8.40 mmol) of sodium cyanate in C_2H_5OH were added to the methanolic solution of cobalt nitrate 1.00g (3.40 mmol) and this was followed by microwave irradiation for few minutes after each addition by using IFB 25 BG-1S model microwave oven. The resulting precipitate was filtered off, washed with 1:1 C_2H_5OH: CH_3OH mixture and dried under vacuum. A dark green colored complex was obtained with the yield of 58.87%.

Instrumentations

Elemental analyses C,H,N were performed using Thermo Finnegan make, Flash EA1112 Series CHNS(O) analyzer. The electrical conductivity measurements were conducted using 10^{-3} M solutions of the metal complex in acetonitrile with Systronic Conductivity Bridge (model number-304) at 30°C. The UV-Visible spectrum of the Co(II), Ni(II) and Cu(II) complexes were recorded on Varian, Cary 5000 model UV-Vis Spectrophotometer. Infrared spectra for the complex and the ligands were recorded on a Perkin Elmer, Spectrum RX-I, FT-IR Spectrometer in KBr discs at room temperature. Thermal analysis of a complexes were measured from room temperature to 900 °C at a heating rate of 10 °C/min in N2 atmosphere using SDT Q 600 V8.3 Build 101. Both TG and DTA were recorded simultaneously.

Antimicrobial activity

The free ligands and their Co(II), Ni(II) and Cu(II) complexes were tested for in vitro antimicrobial activity by the well diffusion method [3], using the agar nutrient as the medium. The antibacterial and the antifungal activities of the ligands and their complexes were evaluated by the well diffusion method against the strains, cultured on potato dextrose agar as medium. In this typical procedure [4], a well was made on the agar medium inoculated with the microorganisms. The well was filled with the test solution using a micropipette and the plate was incubated for 24 hours for bacteria and 72 hours for fungi at 35oC. At the end of the period, the inhibition zones formed on the medium were evaluated as millimeters (mm) diameter.

Antioxidant activity

Assessment of antioxidant activity stock solution (1 mg/ml) was diluted to final concentrations of 10–500 µg/ml. Ethanolic DPPH solution (1 ml, 0.3 mmol) was added to the sample solutions in DMSO (3 ml) at different concentrations (10–500 µg/ml) [5]. The mixture was shaken energetically and acceptable to stand at room temperature for 30 min. The absorbance was then measured at 517 nm in a UV-Vis Spectrophotometer. The lower absorbance of the reaction mixture indicates higher free radical scavenging activity. Ethanol was used as the solvent and ascorbic acid as the standard. The DPPH radical scavenging activity is designed by the following equation:

$$\text{DPPH Scavenging effect (\%)} = \frac{A_o - A_1}{A_o} \times 100$$

where A_o and A_1 are the absorbance of the control reaction and absorbance in the presence of the samples/standard.

DNA binding studies

DNA binding experiments involving the interaction of the synthesized complexes and the ligand with calf thymus CT-DNA were conducted in Tris buffer containing HCl (0.01 M) adjusted to pH 7.2 with hydrochloric acid. The CT-DNA was dissolved in Tris-HCl buffer and was dialyzed against the same buffer overnight. Solutions of CT-DNA gave the ratios of UV absorbance at 260 and 280 nm above 1.8, demonstrating that the DNA was adequately free of protein. DNA concentration per nucleotide was determined by absorption spectroscopy using the molar absorption coefficient 6600 dm3mol-1cm-1 at 260 nm. The stock solutions were stored at 4°C and used within 4 days [6,7]. For fluorescence-quenching experiments, DNA was pre-treated with ethidium bromide (EtBr) for 30 minutes. The prepared complexes then added to this mixture and their effect on the emission intensity was measured. Samples were excited at 450 nm and emission was observed between 500 nm and 800 nm.

RESULTS AND DISCUSSION

Elemental Analysis And Metal Estimation

From the elemental analytical data, the molecular formulae for the metal complexes were determined. It was well consonant with the theoretical values. The values given in parentheses are theoretical, calculated on the basis of the molecular formula of each complex. The experimental data were in good agreement with the theoretical values. (Table 1)

Molar Conductance

Molar conductance measurements of the complexes, carried out using acetonitrile as the solvent at the concentration of 10^{-3}M, indicate non-electrolyte behaviour of the complex [8]. Thus the complexes may be formulated as 1:0 types.

UV-Visible Spectrum of Complexes

The electronic spectrum of Co(II) complex exhibits absorbtion bands at 17241, 18691 and 22727 cm^{-1} and their corresponding transitions [9] may be assigned as $^4T_{1g}(F) \rightarrow {}^4T_{2g}(F)$ (ν_1), $^4T_{1g}(F) \rightarrow {}^4A_{2g}(F)$ (ν_2), $^4T_{1g}(F) \rightarrow {}^4T_{1g}(P)$ (ν_3)which are characteristic of six coordinate octahedral geometry of Co(II) complexes. A moderately intense band seen at 31250 cm^{-1} can be confined to Ligand to metal charge transfer (LMCT) transition. The observed magnetic moment, 4.41 B.M is close to hexa-coordination around the Co(II) metal ion. The electronic spectrum of Ni(II) complex shows absorbtion bands at 15625, 17543 and 23255 cm^{-1} and their corresponding transitions, which are tentatively assigned as $^3A_{2g}(F) \rightarrow {}^3T_{2g}(F)$ (ν_1), $^3A_{2g}(F) \rightarrow {}^3T_{1g}(F)$ (ν_2), $^3A_{2g}(F) \rightarrow {}^3T_{1g}(P)$ (ν_3) respectively [10,11]. The spectrum also shows a band at 26315 cm^{-1} which may be attributed to the Ligand to metal charge transfer. The observed magnetic moment value of Ni(II) complex is 3.10 B.M. This suggests the presence of octahedral environnent around Ni(II) complex. The electronic spectrum of Cu(II) complex exhibits three absorbance bands at 680 nm, 343 nm and 280 nm and their corresponding transitions are $^2A_{1g} \leftarrow {}^2B_{1g}$, $^2B_{2g} \leftarrow {}^2B_{1g}$ and $^2E_g \leftarrow {}^2B_{1g}$ respectively, which indicate octahedral geometry around Cu(II) metal ion. The magnetic moment value of Cu(II) complex is 1.80 B.M, that indicates further confirming hexa-coordination around Cu(II) metal ion [12,13].

FT-IR Spectra of Free Ligands And Their Complexes

The FT-IR spectra of the free ligands and their complexes were recorded in the region of 4000-400 cm^{-1}. The free ligand 4-methylaminopyridine shows in characteristic absorption band at 3327 cm^{-1} assigned as ν(NH) [14]. A small band noticed at 3040 cm^{-1} is due to ν(CH) aryl. Aromatic ν(C= C) stretching vibration is seen as a sharp peak at 1520 cm^{-1} [14,15]. The cyanate ion shows ν(OCN$^-$) 1208 cm^{-1}. The stretching frequency of ν(NH) in 4-methylaminopyridine and the (N$_3^-$) group of the azide ion underwent higher wavenumber at after complexation, indicating the coordination of amino nitrogen and cyanate nitrogen to the metal atom. The low frequency region of the spectra revealed the presence of medium intensity bands in the region of 600-300 cm^{-1} due to ν(M-N) and ν (M-N) [16] respectively in the complexes which supports the involvement of N, N [17] vibrations in complexes which again supports complexation with the metal ion under investigation. Thus, the IR spectral data suggest that the 4-MAP and cyanate ion are bound to the metal ion through the amino nitrogen and cyanate nitrogen donor atoms.

EPR spectrum of Cu(II) complex

The spectrum of DMSO solution of Cu(II) complex of 4-methylaminopyridine (4-MAP) and cyanate ion measured at X-band frequency at 77 K (LNT) provide useful information which is important in studying metal ion environment. The spin Hamiltonian parameters of the complex have been calculated and are summarized. in Table 2. The Cu(II) complex in the frozen state at 77 K shows four well resolved peaks in the low field region and one intense peak in the high field region. The g-tensor value of the copper complex can be used to derive the ground state. In octahedral complexes, the unpaired electron lies in the dx^2-y^2 orbital [18]. For this complex, the observed g-tensor values are $g_\parallel$ =2.2461 > $g_\perp$= 2.2103 > g = 2.0023 which suggest that this complex has an octahedral geometry and the ground state is ^2B_1g. The EPR parameters of the complex coincide well with the related systems which confirm that the complex has an octahedral geometry and it is axially symmetric. In the axial spectra, the g-values are related to the exchange interaction coupling constant G by the expression [19].

$$G = g_\parallel - 2.0023 / g_\perp - 2.0023$$

According to Hathaway [20] expression, if G value is larger than four, the exchange interaction is negligible because the local tetragonal axes are aligned parallel or slightly misaligned. If its value is less than four, the exchange interaction is considerable and the local tetragonal axes are misaligned. For the present Cu(II)complex, G is 0.6241,

which indicates considerable exchange interaction in the solid complex. The g_{av} and the covalent in-plane σ-bonding (α^2) parameters are calculated according to the following equation [21].

$g_{av} = 1/3[g_{\parallel} + 2g_{\perp}]$

$\alpha^2 Cu = (A_{\parallel}/0.036) + g_{\parallel} - 2.0023 + 3/7(g_{\perp} - 2.0023) + 0.04$

If the α^2 value is 0.4142, it indicates a complete covalent bonding, and if the value is 1.0, it suggests a complete ionic bonding. From Table1, it is clear that the in-plane σ -bonding parameter α^2= 0.4142 [32] is less than unity and this indicates the covalent character of M-L bond [22]. These data are well in accordance with the other reported values.

Biological Activity
Antibacterial Activity
The synthesized complexes and the free ligand 4-methylaminopyridine are tested against the bacteria *viz.*, *Escherichia coli*, *Lactobacillus brevis* and *Staphylococcus* by agar-well diffusion method in *vitro* conditions. The complexes have prospective activity against the bacteria compared to free ligand 4-methylaminopyridine.

Antifungal Activity
The antifungal activity of the free ligand 4-methylaminopyridine (4-MAP) and the synthesized complexes are tested against the fungi *viz.*, *Candida albicans, Aspergillus flavus, Aspergillus niger, Aspergillus sojae* and *Aspergillus oryzae* by agar -well diffusion method. The complexes have enhanced activity against the few fungi compared to free ligand 4-methylaminopyridine.

Antioxidant Activity (Radical Scavenging Activity)
Antioxidant activity of the free ligand 4-methylaminopyridine (4-MAP) and the complex were determined by DPPH free radical scavenging method and vitamin C as standard. The reduction capability of DPPH radicals was determined by decrease in its absorbance at 517 nm induced by antioxidants [23]. The graph was plotted with percentage scavenging effects on the y-axis and concentration (µg/ml.) on the x-axis. The scavenging ability of the complexes was compared with Vitamin C as a standard. The metal complexes showed enhance activity as a radical scavenger compared with ascorbic acid, these results were in good agreement with previous metal complexes studies where the ligand has the antioxidant activity and it is expected that the metal moiety will increase its activity [24,25]. The scavenging activities of ligand and their complex shown in Fig.4.

DNA Binding – Emission Study
DNA binding of free ligand and their complexes to CT-DNA can be studied by competitive binding experiments. Ethidium bromide (EtBr) is known to show fluorescence when bound to DNA, due to its strong intercalation between the adjacent DNA base pair. The fluorescent light is quenched by the addition of a second molecule [26,27]. The quenching extent of fluorescence of EtBr binding to DNA is used to determine the extent of binding between the second molecule and DNA. The addition of the complex to DNA pretreated with ethidium bromide causes appreciable reduction in the emission intensity, indicating the replacement of the ethidium bromide fluorophore by the complexes results in a decrease of the binding constant of the ethidium to the DNA as shown in Fig.5 & 6. According to the classical Stern-Volmer equation: Io/I = 1 + Ksvr, where Io and I are the fluorescence intensities in the absence and the presence of complex respectively. Ksv is a linear Stern–Volmer quenching constant, r is the ratio of the total concentration of complex to that of DNA. The quenching plots show that the quenching of ethidium bromide bound to DNA by the complex are in good agreement with the linear Stern-Volmer equation, which also indicates that the complex binds to DNA. In the plot of Io/I versus $C_{Complex}/C_{DNA}$, K is given by the ratio of the slope to intercept and given in Table 3. The obtained Kb values suggest that the interaction of complex with DNA is strongest, which is consistent with the above absorption spectral results. Kb values indicate that the interaction of the complex with DNA is a intercalative mode [28].

CONCLUSION

In the near study, our efforts was to synthesize and characterize a new series of transition metal complexes Co(II), Ni(II) and Cu(II) complexes with 4-methylaminopyridine and cyanate ion as ligands. The new complex was synthesized using microwave irradiation. The synthesized complexes were characterized by various chemical and spectral analyses. The synthesized complex was tested for antimicrobial activities. The metal complex has significant antimicrobial and antioxidant activities as compared to the free ligands. The effectiveness of the DNA binding of the complexes is being confirmed by means of change in intensity of emission in the case of emission spectral studies.

ACKNOWLEDGEMENT

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Table 1 Analytical data of 4-MAP and its metal complexes

S. No	Ligand/complexes	Elements found (Calc) %			Conductance (ohm^{-1}cm^{2}mol^{-1})
		C	H	N	
1	4-Methylaminopyridine	57.15 (51.16)	5.05 (4.11)	21.61 (17.71)	–
2	[Co(OCN)$_2$(4-MAP)$_4$]	51.95 (52.90)	6.27 (5.29)	9.65 (8.67)	74.58
3	[Ni(OCN)$_2$(4-MAP)$_4$]	54.97 (53.93)	6.28 (5.30)	9.63 (9.60)	68.77
4	[Cu(OCN)$_2$(4-MAP)$_4$]	8.63 (9.83)	6.02 (7.22)	9.09 (8.55)	67.65

Table 2. Spin Hamiltonian parameters of Cu(II) complex at 77K

Complex	Spin Hamiltonian parameters					
Cu(II) complex	$g_{\parallel\parallel}$	$g_{\perp}$	g_{av}	G	$A_{\parallel}$10^{-4}cm^{-1}	α^2
	2.2321	2.2101	0.4127	0.6241	151.22	0.4142

Table 3. DNA-binding constant (Kb) of ligand and complexes

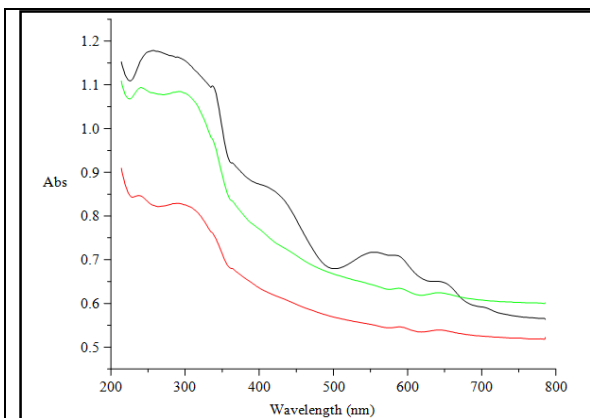| S. No.	Ligand/ Complexes	Binding constant (Kb)
1	4-Methylaminopyridine	1.74 × 10^4 M^{-1}
2	[Co(OCN)$_2$(4-MAP)$_4$]	3.23 × 10^4 M^{-1}
3	[Ni(OCN)$_2$(4-MAP)$_4$]	2.17 × 10^4 M^{-1}
4	[Cu(OCN)$_2$(4-MAP)$_4$]	3.41 × 10^4 M^{-1}

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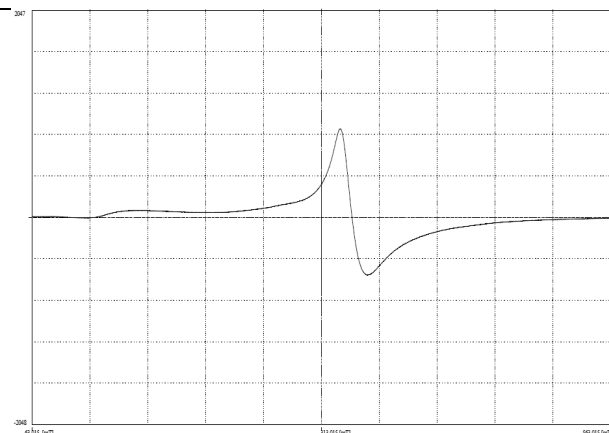**Fig.1 Electronic spectra of Co(II), Ni(II) and Cu(II) complexes**

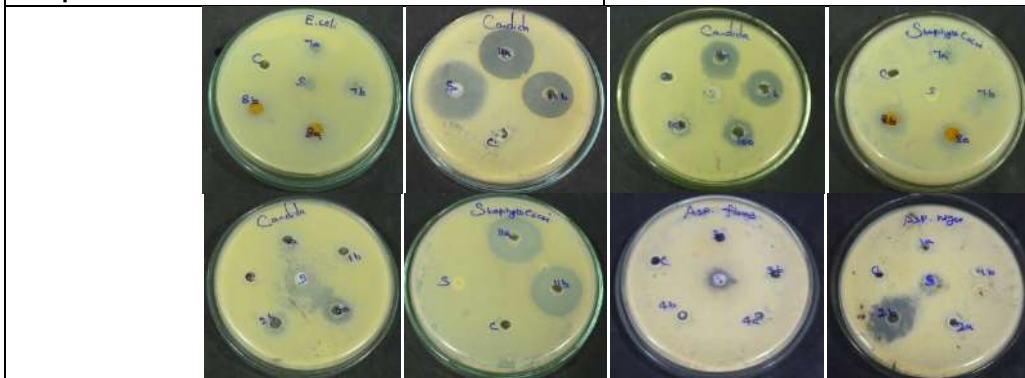**Fig.2 EPR spectrum of Cu(II) complex**
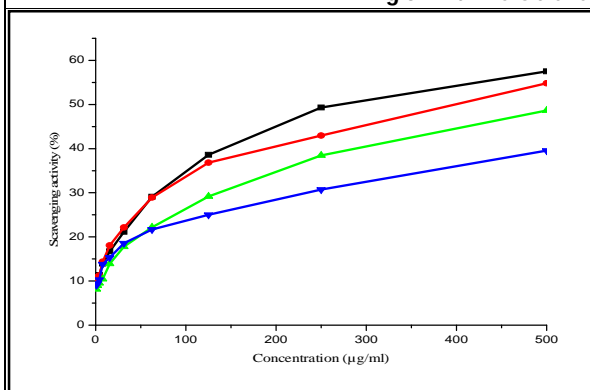

Fig.3 Antimicrobial activities of Co(II) complex

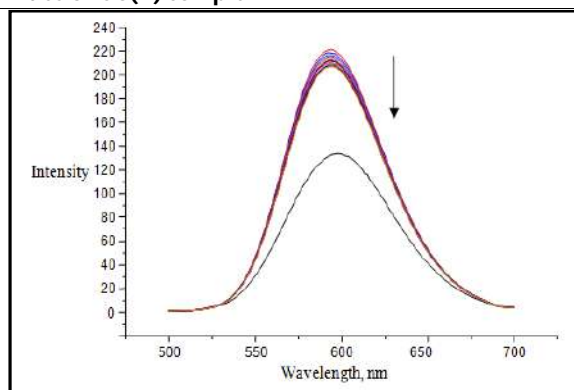**Fig.4 Antioxidant activities of 4-MAP,Co(II), Ni(II) and Cu(II) complexes**

Fig 5. Emission spectrum of 4- methylaminopyridine

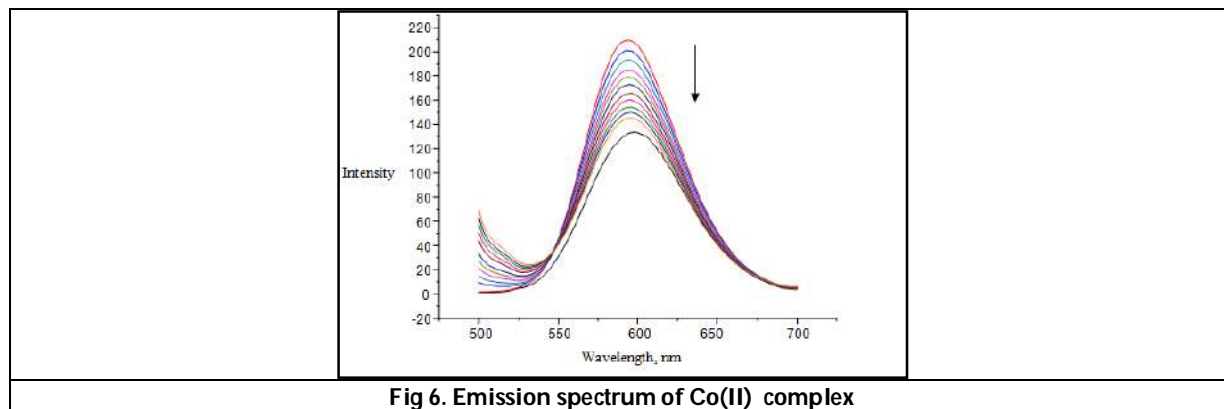**Fig 6. Emission spectrum of Co(II) complex**

RESEARCH ARTICLE

Study the Effective of Earthquake Load on Wall Pier Forces by Linear Dynamics Analysis

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ABSTRACT

A wall pier is a segment of a wall whose horizontal length is between two and one-half and six times its thickness whose clear height is at least two times its horizontal length, wall segments are components that are longer than wall piers. They are the primary resisting components in the shear wall. In the present Research work G+29 story building with different type of RC Shear wall at the center in Concrete Frame Structure with fixed support conditions under different type of soil for high seismic zone are analyzed. This paper aims to study the effect of seismic load on wall pier forces in different type of RC shear walls in concrete frame structures under different type of soil condition. Estimation of pier forces such as; wall pier axial force, wall pier moment, wall pier shear force, wall pier torsion, time period and frequency and modal load participation ratios is carried out. In dynamic analysis; response spectrum method with ETABS software is used. It was found that the behavior of new shape (plus shape) of RC shear wall are not more different with I and box shape and .It was found that the tall building with box shape Shear Walls provided at the center core of structure showed better performance in terms of wall pier forces, time period and frequency.

Keywords: Linear Dynamics Analysis; Wall Pier Forces; Reinforced Concrete Shear Wall ; High Seismic Zone

INTRODUCTION

Structural Systems

In the earliest structures at the beginning of the 20th century, structural members were assumed to carry primarily the gravity loads. Today, however, by the advances in structural design / systems and high-strength materials, building height is increased, which necessitates taking into consideration mainly the lateral loads such as wind and earthquake. Understandably, especially for the tall buildings, as the slenderness, and so the flexibility increases, buildings suffer from the lateral loads resulting from wind and earthquake more and more. As a general rule, when other things being equal, the taller the building, the more necessary it is to identify the proper structural system for resisting the lateral loads. Currently, there are many structural systems that can be used for the lateral resistance of tall buildings [2,3]. Structural systems of tall buildings can be divided into two broad categories: interior structures and exterior structures. This classification is based on the distribution of the components of the primary lateral load-resisting system over the building.

Shear wall structure

Shear Wall–Frame Systems (Dual Systems), The system consists of reinforced concrete frames interacting with reinforced concrete shear walls are adequate for resisting both the vertical and the horizontal loads acting on them.

Necessity of Shear Walls

Shear wall system has two distinct advantages over a frame system.
- It provides adequate strength to resist large lateral loads with-out excessive additional cost.
- It provides adequate stiffness to resist lateral displacements to permissible limits, thus reducing risk of non-structural damage.

Earthquake Load

The seismic weight of building is the sum of seismic weight of all the floors [8]. The seismic weight of each floor is its full dead load plus appropriate amount of imposed load, the latter being that part of the imposed loads that may reasonably be expected to be attached to the structure at the time of earthquake shaking. Earthquake forces experienced by a building result from ground motions (accelerations) which are also fluctuating or dynamic in nature, in fact they reverse direction somewhat chaotically[2,3]. In theory and practice, the lateral force that a building experiences from an earthquake increases in direct proportion with the acceleration of ground motion at the building site and the mass of the building . As the ground accelerates back and forth during an earthquake it imparts back-and-forth (cyclic) forces to a building through its foundation which is forced to move with the ground [1].

Geo-Technical Consideration

The seismic motion that reaches a structure on the surface of the earth is influenced by local soil conditions. The subsurface soil layers underlying the building foundation may amplify the response of the building to earthquake motions originating in the bedrock. Three soil types are considered here: Hard, Medium & Soft soils ,the allowable bearing pressure shall be determined in accordance with IS: 1888-1982 load test (Revision 1992).

METHODOLOGY

a) To understand and evaluation building structures and aims to the effect of Seismic load on Wall Pier Forces in Different Type of RC Shear Walls in Concrete Frame Structures under Different Type of Soil Condition with seismic loading.
b) Modeling a G+29 story high building for five different cases [9-10,11].
c) Analyzing the building dynamic analysis using linear ,i.e. Response Spectrum Analysis [1-2,3].
d) Analyzing the results and arriving at conclusions.

Dynamic analysis
Dynamic analysis may be executed to get the design seismic force, and its spread in different levels through the height of the building, and also various lateral load resisting element [1-2-3,8].

Response Spectrum Method
This method is executed to design spectrum, where as it is specified with a code for specific- site design can be used for a project site for the purposes of dynamic of steel and reinforce concrete buildings, the values of damping for building may be taken as 2 and 5 percent of the critical, respectively. response spectrum method is typically implemented in linear elastic procedures and also very much easier to use This also called as or mode superposition method or model method, It also made on the idea of the superposition of responses given by the building through various modes of vibrations, each vibration modes is recorded as with its own particular deformed shape, with its own modal damping and its own frequency [7,8].

MODELING OF BUILDING

Details of the Building
A symmetrical building [15] of plan 38.5m X 35.5m located with location in high Seismic zone considered. Four bays of length 7.5m& one bays of length 8.5m along X - direction and four bays of length 7.5m& one bays of length 5.5m along Y - direction are provided. Shear is provided the center inner core of model building.
Struct I: G+29 story's tall building with Plus shape RC shear wall at the center of structure .
Struct II: G+29 story's tall building with Box shape RC shear wall at the center of structure .
Struct III: G+29 story's tall building with C- shape RC shear wall at the center of structure .
Struct IV: G+29 story's tall building with E- shape RC shear wall at the center of structure .
Struct V: G+29 story's tall building with I- shape RC shear wall at the center of structure .

Load Combinations
As per IS 1893 (Part 1): 2002 Clause no. 6.3.1.2, the following load cases have to be considered for analysis:
"1.2 (DL + IL ± EL)"
"1.5 (DL ± EL)"
"EQXP&EQYP"
Earthquake load must be considered for +X, -X, +Y and –Y Directions [5-6,7].

The Building Details
Type of frame: Special RC moment resisting frame fixed at the base, Number of storeys: G+29, Floor height:3.5 m , Depth of Slab:225 mm , Size of beam:(300 × 600) mm, Size of column (exterior) :(1250×1250) mm up to story five , Size of column (exterior): (900×900) mm Above story five , Size of column (interior) :(1250×1250) mm up to story ten, Size of column (interior): (900×900) mm Above story ten, Live load on floor:4 KN/m2, Floor finish:2.5 KN/m2, Wall load:25 KN/m, Grade of Concrete: M 50 concrete, Grade of Steel: Fe 500, Thickness of shear wall:450 mm , Seismic zone: V, Important Factor:1.5, Density of concrete:25 KN/m3, Type of soil: Type I=Soft Soil, Type II=Medium Soil, Type III= Hard Soil, Response spectra: As per IS 1893(Part-1):2002,Damping of structure:5 percent& All the analyses has been carried out as per the Indian Standard code books [4-5-6-7,8].

RESULTS AND DISCUSSIONS

At each section along the height, shear wall carries[16],
- Axial Force P
- Shear Force V
- Bending Moment M

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Parametric Results in Wall Pier Axial Force, Wall Pier Moment, Wall Pier Shear Force & Wall Pier Torsion with Different load combination in Different Type of Soil Condition were considered ,compared all wall pier Forces in different type of soil condition of Structures II,III,IV,V with Structure I.

DISCUSSION ON RESULTS

When a structure is subjected to earthquake, it responds by vibrating. An example force can be resolved into three mutually perpendicular directions- two horizontal directions (X and Y directions) and the vertical direction (Z) [8]. This motion causes the structure to vibrate or shake in all three directions; the predominant direction of shaking is horizontal. All the structures are primarily designed for gravity loads-force equal to mass time's gravity in the vertical direction. Vertical acceleration should also be considered in structures with large spans those in which stability for design, or for overall stability analysis of structures. The basic intent of design theory for earthquake resistant structures is that buildings should be able to resist minor earthquakes without damage, resist moderate earthquakes without structural damage but with some non-structural damage. To avoid collapse during a major earthquake, Members must be ductile enough to absorb and dissipate energy by post elastic deformation. Redundancy in the structural system permits redistribution of internal forces in the event of the failure of key elements. When the primary element or system yields or fails, the lateral force can be redistributed to a secondary system to prevent progressive failure.

When a structure is subjected to an earthquake excitation, it interacts with the foundation and the soil, and thus changes the motion of the ground[2,8]. This means that the movement of the whole ground-structure system is influenced by the type of soil as well as by the type of structure. Understanding of soil structure interaction will enable the designer to design structures that will behave better during an earthquake.

CONCLUSIONS

From the above results and discussions, following conclusions can be drawn:
- It was found that the behavior of Tall building with new shape (Plus Shape) RC shear wall are not more different with I and box shape RC shear wall .
- The shear wall and it is position has a significant influenced on the time period. The time period is not influenced by the type of soil. In Tall Building with box shape Shear Walls is showing the low time period which shows a very significant performance.
- There is considerable difference in Pier Moment with a Different type of soils and structures.
- There is considerable difference in Pier shear force with a Different type of soils and structures.
- There is not considerable difference in Pier axial forces with a Different type of soils and structures.
- It is evident that the Pier Shear of medium soil and hard soil with soft soil for Tall building with Box Shape RC shear wall is similar with the Tall building with C- Shape RC shear wall .
- It is evident that the Pier Torsion of medium soil and hard soil with soft soil for Tall building with Box Shape RC shear wall is similar with the Tall buildings with C&I Shape RC shear wall .
- The Pier Axial Forces for the Load Combinations in Y direction showed the same performance in all type of soil .
- The Pier Moment for the Load Combinations in Y direction showed the same performance in all type of soil .
- It is evident that Pier Torsion in X direction for all structures in soft soil more than Medium soil and more than hard soil.
- It is evident that Pier Torsion in Y direction for soft soil less than Medium soil and less than hard soil.
- shear is effected marginally by placing of the shear wall, grouping of shear wall and type of soil. The shear is increased by adding shear wall due to increase the seismic weight of the building.
- The moment resisting frame with shear walls are very good in lateral force such as earthquake and wind force. The shear walls provide lateral load   distribution by transferring the wind and   earthquake loads to the foundation. And also impact on the lateral stiffness of the system and also carries gravity loads.

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- According to IS-1893:2002 the number of modes to be used in the analysis should be such that the total sum of modal masses of all modes considered is at least 90 percent of the total seismic mass. Here the maximum mass is for the tall building with Box Shape RC shear wall.
- ETABS is the robust software which is utilized for analysing any kind of multi building structures.

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Table 1.Wall Pier Axial Force, P for structures with the load combination 1.5 (DL +EQXP) &1.5 (DL +EQYP) , All value in "kN"

Wall Pier Axial Force, P in soft soil								
Pier Forces				Struct I	Struct II	Struct III	Struct IV	Struct V
Story	Pier	Load Case/Combo	Location	"P"	"P"	"P"	"P"	"P"
1ST	P3	1.5(DL+EQXP)	Top	-30667.2385	-34801.7254	-33838.7116	-4425.5079	-32394.3369
1ST	P3	1.5(DL+EQXP)	Bottom	-30992.0822	-35126.5691	-34163.5554	-4587.9297	-32719.1806
1ST	P3	1.5(DL+EQYP)	Top	-30667.2385	-24951.1394	-31627.4224	-13342.3348	-32953.4102
1ST	P3	1.5(DL+EQYP)	Bottom	-30992.0822	-25275.9831	-31952.2661	-13504.7567	-33278.2539

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Wall Pier Axial Force, P in medium soil							
1ST	P3	1.5(DL+EQXP)	Top	-30667.2385	-38347.9363	-34634.7757	-466.9035	-32193.1202
1ST	P3	1.5(DL+EQXP)	Bottom	-30992.0822	-38672.7801	-34959.6195	-629.3254	-32517.9639
1ST	P3	1.5(DL+EQYP)	Top	-30667.2385	-24951.1394	-31627.4224	-13342.3348	-32953.4598
1ST	P3	1.5(DL+EQYP)	Bottom	-30992.0822	-25275.9831	-31952.2661	-13504.7567	-33278.3036
Wall Pier Axial Force, P in hard soil							
1ST	P3	1.5(DL+EQXP)	Top	-30667.2385	-41401.618	-35320.2754	2467.9374	-32019.8502
1ST	P3	1.5(DL+EQXP)	Bottom	-30992.0822	-41726.4617	-35645.1192	2305.5156	-32344.694
1ST	P3	1.5(DL+EQYP)	Top	-30667.2385	-24951.1394	-31627.4224	-13342.3348	-32953.5026
1ST	P3	1.5(DL+EQYP)	Bottom	-30992.0822	-25275.9831	-31952.2661	-13504.7567	-33278.3463

Table 2.Wall Pier Moment, M for structures with the load combination1.5 (DL +EQXP) &1.5 (DL +EQYP) , All value in "kN-m"

Wall Pier Moment, M in soft soil													
TABLE: Pier Forces				Struct I	Struct I	Struct II	Struct II	Struct III	Struct III	Struct IV	Struct IV	Struct V	Struct V
Story	Pier	Load Case/Combo	Location	"M2"	"M3"	"M2"	"M3"	"M2"	"M3"	"M2"	"M3"	"M2"	"M3"
1ST	P3	1.5(DL+EQXP)	Top	540.9001	-239.0392	67.293	1500.4694	0.9565	297.2489	-21.8361	2510.6932	-63.8605	-76.9109
1ST	P3	1.5(DL+EQXP)	Bottom	-997.463	-204.4984	-553.8033	-456.3434	-619.2684	504.0265	26.5312	7894.7587	-380.7271	-21.7227
1ST	P3	1.5(DL+EQYP)	Top	-8.6497	36985.9055	0.4472	23858.1637	-0.2579	23522.4129	26.8211	-711.4265	-54.7972	13884.4054
1ST	P3	1.5(DL+EQYP)	Bottom	3.4013	51592.3539	-3.2036	34967.344	-4.5199	39534.6011	173.4753	1020.3153	62.739	43569.6278
Wall Pier Moment, M in medium soil													
1ST	P3	1.5(DL+EQXP)	Top	738.738	-239.0392	91.3574	2040.6384	1.3937	404.2585	-21.8361	2510.6932	-67.1221	-89.0173
1ST	P3	1.5(DL+EQXP)	Bottom	-1357.7742	-204.4984	-752.0192	-620.6271	-840.5778	685.4761	26.5312	7894.7587	-540.3743	-20.8092
1ST	P3	1.5(DL+EQYP)	Top	-8.6497	50386.8856	0.4472	32447.1026	-0.2579	31990.4816	38.7433	-711.4265	-54.796	18898.3728
1ST	P3	1.5(DL+EQYP)	Bottom	3.4013	70239.2207	-3.2036	47555.5879	-4.5199	53767.0575	250.4668	1020.3153	62.7395	59263.4276
Wall Pier Moment, M in hard soil													
1ST	P3	1.5(DL+EQXP)	Top	909.0984	-239.0392	112.0796	2505.7839	1.7701	496.4057	-38.7129	5001.4843	-69.9307	-99.4423
1ST	P3	1.5(DL+EQXP)	Bottom	-1668.0421	-204.4984	-922.7051	-762.0936	-1031.1499	841.7243	47.0368	13208.1901	-677.8482	-20.0225
1ST	P3	1.5(DL+EQYP)	Top	909.0984	-239.0392	0.4472	39843.1333	-0.2579	39282.4296	47.5745	-711.4265	-54.795	23215.9559
1ST	P3	1.5(DL+EQYP)	Bottom	-1668.0421	-204.4984	-3.2036	58395.4646	-4.5199	66022.7839	307.5585	1020.3153	62.74	72777.5329

Table 3.Wall Pier Shear Force, V for structures with the load combination 1.5 (DL +EQXP) &1.5 (DL +EQYP),All value in "kN"

Wall Pier Shear Force, V in soft soil													
Pier Forces				Struct I	Struct I	Struct II	Struct II	Struct III	Struct III	Struct IV	Struct IV	Struct V	Struct V
Story	Pier	Load Case/Combo	Location	"V2"	"V3"	"V2"	"V3"	"V2"	"V3"	"V2"	"V3"	"V2"	"V3"
1ST	P3	1.5(DL+EQXP)	Top	9.8688	-439.5323	-559.0894	-177.4561	59.0793	-177.2071	1538.3044	13.8192	15.768	-90.5333
1ST	P3	1.5(DL+EQXP)	Bottom	9.8688	-439.5323	-559.0894	-177.4561	59.0793	-177.2071	1538.3044	13.8192	15.768	-90.5333
1ST	P3	1.5(DL+EQYP)	Top	4173.271	3.4431	3174.0515	-1.0431	4574.9109	-1.2177	494.7834	41.9012	8481.4921	33.5818
1ST	P3	1.5(DL+EQYP)	Bottom	4173.271	3.4431	3174.0515	-1.0431	4574.9109	-1.2177	494.7834	41.9012	8481.4921	33.5818
Wall Pier Shear Force, V in medium soil													
1ST	P3	1.5(DL+EQXP)	Top	9.8688	-599.0035	-760.3616	-240.9648	80.3479	-240.5633	2001.3618	19.952	19.488	-135.2149
1ST	P3	1.5(DL+EQXP)	Bottom	9.8688	-599.0035	-760.3616	-240.9648	80.3479	-240.5633	2001.3618	19.952	19.488	-135.2149
1ST	P3	1.5(DL+EQYP)	Top	5672.0958	3.4431	4316.7101	-1.0431	6221.8788	-1.2177	494.7834	60.4924	11532.8728	33.5816
1ST	P3	1.5(DL+EQYP)	Bottom	5672.0958	3.4431	4316.7101	-1.0431	6221.8788	-1.2177	494.7834	60.4924	11532.8728	33.5816
Wall Pier Shear Force, V in hard soil													
1ST	P3	1.5(DL+EQXP)	Top	9.8688	-736.3259	-933.6793	-295.6528	98.6625	-295.12	2344.7731	24.4999	22.6914	-173.6907
1ST	P3	1.5(DL+EQXP)	Bottom	9.8688	-736.3259	-933.6793	-295.6528	98.6625	-295.12	2344.7731	24.4999	22.6914	-173.6907
1ST	P3	1.5(DL+EQYP)	Top	9.8688	-736.3259	5300.6661	-1.0431	7640.1012	-1.2177	494.7834	74.2811	14160.4506	33.5814
1ST	P3	1.5(DL+EQYP)	Bottom	9.8688	-736.3259	5300.6661	-1.0431	7640.1012	-1.2177	494.7834	74.2811	14160.4506	33.5814

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Table 4. Wall Pier Torsion, T for structures with the load combination 1.5 (DL +EQXP) &1.5 (DL +EQYP), All value in "kN"

Wall Pier Torsion, T in soft soil				Struct I	Struct II	Struct III	Struct IV	Struct V
TABLE: Pier Forces				"P"	"P"	"P"	"P"	"P"
Story	Pier	Load Case/Combo	Location	"P"	"P"	"P"	"P"	"P"
1ST	P3	1.5(DL+EQXP)	Top	-69.1502	-39.7786	-40.3244	-21.6394	-42.4392
1ST	P3	1.5(DL+EQXP)	Bottom	-69.1502	-39.7786	-40.3244	-21.6394	-42.4392
1ST	P3	1.5(DL+EQYP)	Top	61.4016	52.394	116.1891	44.8766	106.4507
1ST	P3	1.5(DL+EQYP)	Bottom	61.4016	52.394	116.1891	44.8766	106.4507
Wall Pier Torsion, T in medium soil								
1ST	P3	1.5(DL+EQXP)	Top	-91.6969	-54.0989	-54.8412	-31.2427	-57.7158
1ST	P3	1.5(DL+EQXP)	Bottom	-91.6969	-54.0989	-54.8412	-31.2427	-57.7158
1ST	P3	1.5(DL+EQYP)	Top	85.8536	71.2559	158.0172	64.792	144.7745
1ST	P3	1.5(DL+EQYP)	Bottom	85.8536	71.2559	158.0172	64.792	144.7745
Wall Pier Torsion, T in hard soil								
1ST	P3	1.5(DL+EQXP)	Top	-111.112	-66.4303	-67.3418	-38.3642	-70.8707
1ST	P3	1.5(DL+EQXP)	Bottom	-111.112	-66.4303	-67.3418	-38.3642	-70.8707
1ST	P3	1.5(DL+EQYP)	Top	-111.112	87.498	194.0359	79.5608	177.7754
1ST	P3	1.5(DL+EQYP)	Bottom	-111.112	87.498	194.0359	79.5608	177.7754

Table 5. Modal Load Participation Ratios

Modal Load Participation Ratios			Struct I	Struct I	Struct II	Struct II	Struct III	Struct III	Struct IV	Struct IV	Struct V	Struct V
Case	Item Type	Item	Static	Dynamic	Static	Dynamic	Static	Dynamic	Static	Dynamic	Static	Dynamic
			%	%	%	%	%	%	%	%	%	%
Modal	Acceleration	UX	99.82	86.71	99.99	94.7	99.98	94.59	99.99	94.54	99.97	91.54
Modal	Acceleration	UY	99.79	87.46	99.98	91.46	99.97	91.85	99.97	91.83	99.97	92.51
Modal	Acceleration	UZ	0	0	0	0	0	0	0	0	0	0

Table 6. Modal Periods and Frequencies

		Struct I		Struct II		Struct III		Struct IV		Struct V	
Case	Mode	Period	Frequency	Period	Frequency	Period	Frequency	Period	Frequency	Period	Frequency
		sec	cyc/sec	sec	cyc/sec	sec	cyc/sec	sec	cyc/sec	sec	cyc/sec
Modal	1	6.298	0.159	5.785	0.173	6.415	0.156	6.375	0.157	6.382	0.157
Modal	2	6.248	0.16	5.606	0.178	6.32	0.158	6.21	0.161	5.694	0.176
Modal	3	5.545	0.18	4.684	0.213	5.767	0.173	5.792	0.173	5.642	0.177
Modal	4	2.062	0.485	1.701	0.588	2.114	0.473	2.102	0.476	2.088	0.479
Modal	5	1.952	0.512	1.547	0.646	1.958	0.511	1.901	0.526	1.565	0.639
Modal	6	1.603	0.624	1.475	0.678	1.568	0.638	1.575	0.635	1.524	0.656
Modal	7	1.191	0.84	0.9	1.112	1.219	0.82	1.212	0.825	1.19	0.84
Modal	8	1.027	0.974	0.838	1.193	1.028	0.972	0.983	1.017	0.791	1.264
Modal	9	0.803	1.245	0.645	1.551	0.82	1.22	0.815	1.226	0.711	1.406
Modal	10	0.782	1.279	0.613	1.632	0.711	1.406	0.714	1.401	0.703	1.423
Modal	11	0.645	1.55	0.5	2.002	0.641	1.56	0.604	1.656	0.565	1.769
Modal	12	0.581	1.72	0.45	2.222	0.592	1.689	0.589	1.697	0.423	2.363

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Table 7. Compared of Wall Pier Axial Forces in Soft soil of Structures II, III, IV, V with Structure I

Pier Forces				Struct II	Struct III	Struct IV	Struct V
Story	Pier	Load Case/Combo	Location	"P"	"P"	"P"	"P"
1ST	P3	1.2(DL+LL+EQXP)	Top & Bottom	4%	8%	-385%	5%
1ST	P3	1.2(DL+LL+EQYP)	Top & Bottom	-26%	3%	-133%	6%
1ST	P3	1.5(DL+EQXP)	Top & Bottom	12%	9%	-584%	5%
1ST	P3	1.5(DL+EQYP)	Top & Bottom	-23%	3%	-130%	7%
1ST	P3	EQXP	Top & Bottom	100%	100%	100%	100%
1ST	P3	EQYP	Top & Bottom	0%	0%	0%	100%

Table 8. Compared of Wall Pier Axial Forces in medium soil of Structures II, III, IV, V with Structure I

Pier Forces				Struct II	Struct III	Struct IV	Struct V
Story	Pier	Load Case/Combo	Location	"P"	"P"	"P"	"P"
1ST	P3	1.2(DL+LL+EQXP)	Top & Bottom	12%	10%	-838%	5%
1ST	P3	1.2(DL+LL+EQYP)	Top & Bottom	-26%	3%	-133%	6%
1ST	P3	1.5(DL+EQXP)	Top & Bottom	20%	11%	-5646%	5%
1ST	P3	1.5(DL+EQYP)	Top & Bottom	-23%	3%	-130%	7%
1ST	P3	EQXP	Top & Bottom	100%	100%	100%	100%
1ST	P3	EQYP	Top & Bottom	0%	0%	0%	100%

Table 9.Compared of Wall Pier Axial Forces in hard soil of Structures II, III, IV, V with Structure I

Pier Forces				Struct II	Struct III	Struct IV	Struct V
Story	Pier	Load Case/Combo	Location	"P"	"P"	"P"	"P"
1ST	P3	1.2(DL+LL+EQXP)	Top & Bottom	17%	11%	-2950%	4%
1ST	P3	1.2(DL+LL+EQYP)	Top & Bottom	-26%	3%	-133%	6%
1ST	P3	1.5(DL+EQXP)	Top & Bottom	26%	13%	1393%	4%
1ST	P3	1.5(DL+EQYP)	Top & Bottom	-23%	3%	-130%	7%
1ST	P3	EQXP	Top & Bottom	100%	100%	100%	100%
1ST	P3	EQYP	Top & Bottom	0%	0%	0%	100%

Table 10.Compared of Wall Pier moment in Soft soil of Structures II,III,IV,V with Structure I

Pier Forces				Struct II	Struct III	Struct IV	Struct V
Story	Pier	Load Case/Combo	Location	"M"	"M"	"M"	"M"
1ST	P3	1.2(DL+LL+EQXP)	Top & Bottom	-384%	-11048%	3205%	293%
1ST	P3	1.2(DL+LL+EQYP)	Top & Bottom	675%	675%	123%	88%
1ST	P3	1.5(DL+EQXP)	Top & Bottom	-392%	-28256%	3218%	393%
1ST	P3	1.5(DL+EQYP)	Top & Bottom	1120%	-1539%	115%	89%
1ST	P3	EQXP	Top & Bottom	-402%	-22608%	3245%	3020%
1ST	P3	EQYP	Top & Bottom	0%	0%	100%	100%

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Table 11.Compared of Wall Pier moment in medium soil of Structures II,III,IV,V with Structure I

Pier Forces				Struct II	Struct III	Struct IV	Struct V
Story	Pier	Load Case/Combo	Location	"M"	"M"	"M"	"M"
1ST	P3	1.2(DL+LL+EQXP)	Top & Bottom	-389%	-12815%	3035%	405%
1ST	P3	1.2(DL+LL+EQYP)	Top & Bottom	675%	675%	116%	88%
1ST	P3	1.5(DL+EQXP)	Top & Bottom	-395%	-26484%	4350%	525%
1ST	P3	1.5(DL+EQYP)	Top & Bottom	1120%	-1539%	110%	89%
1ST	P3	EQXP	Top & Bottom	-402%	-22609%	3062%	3020%
1ST	P3	EQYP	Top & Bottom	0%	0%	100%	100%

Table 12.Compared of Wall Pier moment in hard soil of Structures II,III,IV,V with Structure I

Pier Forces				Struct II	Struct III	Struct IV	Struct V
Story	Pier	Load Case/Combo	Location	"M"	"M"	"M"	"M"
1ST	P3	1.2(DL+LL+EQXP)	Top & Bottom	-391%	-13953%	3643%	492%
1ST	P3	1.2(DL+LL+EQYP)	Top & Bottom	675%	675%	113%	88%
1ST	P3	1.5(DL+EQXP)	Top & Bottom	-396%	-25660%	3047%	627%
1ST	P3	1.5(DL+EQYP)	Top &Bottom	-127577%	157898%	-584%	2259%
1ST	P3	EQXP	Top & Bottom	-402%	-22608%	3062%	3020%
1ST	P3	EQYP	Top & Bottom	0%	0%	100%	100%

Table 13.Compared of Wall Pier shear in Soft soil of Structures II,III,IV,V with Structure I

Pier Forces				Struct II	Struct III	Struct IV	Struct V
Story	Pier	Load Case/Combo	Location	"V"	"V"	"V"	"V"
1ST	P3	1.2(DL+LL+EQXP)	Top & Bottom	103%	75%	99%	8%
1ST	P3	1.2(DL+LL+EQYP)	Top & Bottom	-32%	9%	-569%	51%
1ST	P3	1.5(DL+EQXP)	Top & Bottom	102%	83%	99%	37%
1ST	P3	1.5(DL+EQYP)	Top & Bottom	-31%	9%	-743%	51%
1ST	P3	EQXP	Top & Bottom	100%	100%	100%	100%
1ST	P3	EQYP	Top & Bottom	-31%	9%	0%	51%

Table 14.Compared of Wall Pier shear in medium soil of Structures II, III, IV, V with Structure I

Pier Forces				Struct II	Struct III	Struct IV	Struct V
Story	Pier	Load Case/Combo	Location	"V"	"V"	"V"	"V"
1ST	P3	1.2(DL+LL+EQXP)	Top & Bottom	98%	81%	99%	26%
1ST	P3	1.2(DL+LL+EQYP)	Top & Bottom	-32%	9%	-809%	51%
1ST	P3	1.5(DL+EQXP)	Top & Bottom	101%	88%	100%	49%
1ST	P3	1.5(DL+EQYP)	Top & Bottom	-31%	9%	-1046%	51%
1ST	P3	EQXP	Top & Bottom	-31%	9%	0%	51%
1ST	P3	EQYP	Top & Bottom	100%	100%	100%	100%

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Table 15.Compared of Wall Pier shear in hard soil of Structures II, III, IV, V with Structure I

Pier Forces				Struct II	Struct III	Struct IV	Struct V
Story	Pier	Load Case/Combo	Location	"V"	"V"	"V"	"V"
1ST	P3	1.2(DL+LL+EQXP)	Top & Bottom	102%	85%	99%	36%
1ST	P3	1.2(DL+LL+EQYP)	Top & Bottom	-31%	9%	-1016%	51%
1ST	P3	1.5(DL+EQXP)	Top & Bottom	101%	90%	100%	57%
1ST	P3	1.5(DL+EQYP)	Top & Bottom	100%	100%	98%	100%
1ST	P3	EQXP	Top & Bottom	100%	100%	100%	100%
1ST	P3	EQYP	Top & Bottom	-31%	9%	0%	51%

Table 16.Compared of Wall Pier Torsion in Soft soil of Structures II, III, IV, V with Structure I

Pier Forces				Struct II	Struct III	Struct IV	Struct V
Story	Pier	Load Case/Combo	Location	"T"	"T"	"T"	"T"
1ST	P3	1.2(DL+LL+EQXP)	Top & Bottom	-82%	-79%	-234%	-70%
1ST	P3	1.2(DL+LL+EQYP)	Top & Bottom	-11%	50%	-30%	45%
1ST	P3	1.5(DL+EQXP)	Top & Bottom	-74%	-71%	-220%	-63%
1ST	P3	1.5(DL+EQYP)	Top & Bottom	-17%	47%	-37%	42%
1ST	P3	EQXP	Top & Bottom	-57%	-55%	-189%	-48%
1ST	P3	EQYP	Top & Bottom	-30%	42%	-43%	36%

Table 17.Compared of Wall Pier Torsion in medium soil of Structures II, III, IV, V with Structure I

Pier Forces				Struct II	Struct III	Struct IV	Struct V
Story	Pier	Load Case/Combo	Location	"T"	"T"	"T"	"T"
1ST	P3	1.2(DL+LL+EQXP)	Top & Bottom	-75%	-73%	-204%	-64%
1ST	P3	1.2(DL+LL+EQYP)	Top & Bottom	-16%	48%	-28%	43%
1ST	P3	1.5(DL+EQXP)	Top & Bottom	-69%	-67%	-193%	-59%
1ST	P3	1.5(DL+EQYP)	Top & Bottom	-20%	46%	-33%	41%
1ST	P3	EQXP	Top & Bottom	-30%	42%	-51%	36%
1ST	P3	EQYP	Top & Bottom	-57%	-55%	-173%	-48%

Table 18.Compared of Wall Pier Torsion in hard soil of Structures II, III, IV, V with Structure I

Pier Forces				Struct II	Struct III	Struct IV	Struct V
Story	Pier	Load Case/Combo	Location	"T"	"T"	"T"	"T"
1ST	P3	1.2(DL+LL+EQXP)	Top & Bottom	-72%	-70%	-198%	-61%
1ST	P3	1.2(DL+LL+EQYP)	Top & Bottom	-19%	47%	-30%	42%
1ST	P3	1.5(DL+EQXP)	Top & Bottom	-67%	-65%	-190%	-57%
1ST	P3	1.5(DL+EQYP)	Top & Bottom	227%	157%	240%	163%
1ST	P3	EQXP	Top & Bottom	-57%	-55%	-173%	-48%
1ST	P3	EQYP	Top & Bottom	-30%	42%	-43%	36%

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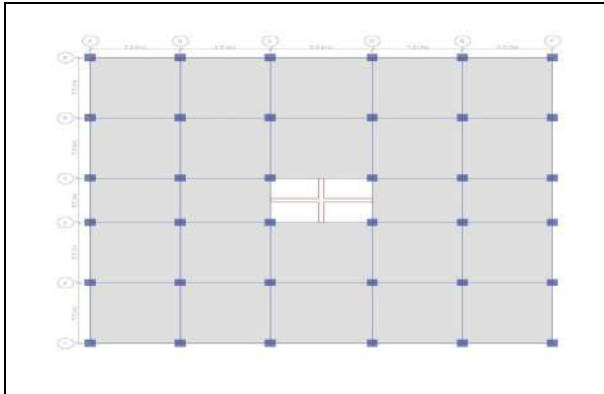
Figure 1. Plan of the Structure I
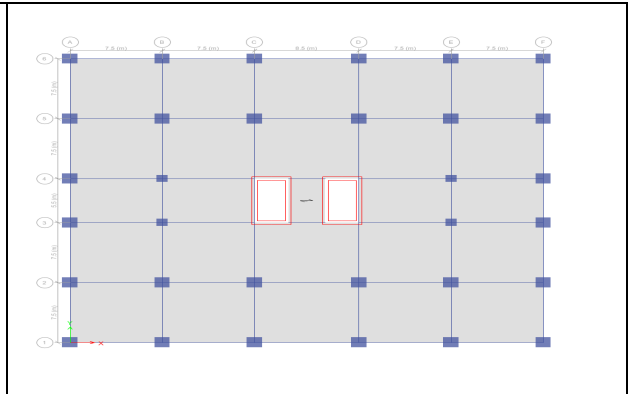
Figure 2. Plan of the Structure II
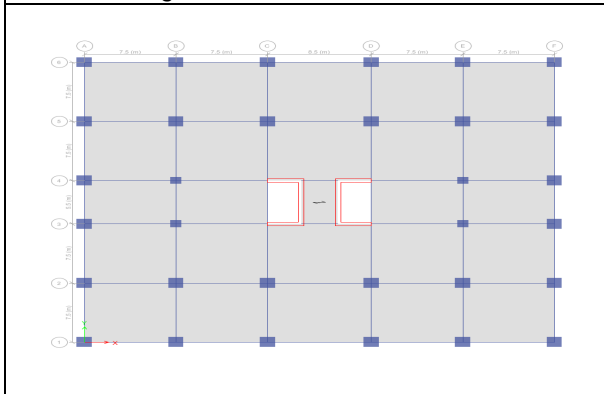
Figure 3. Plan of the Structure III
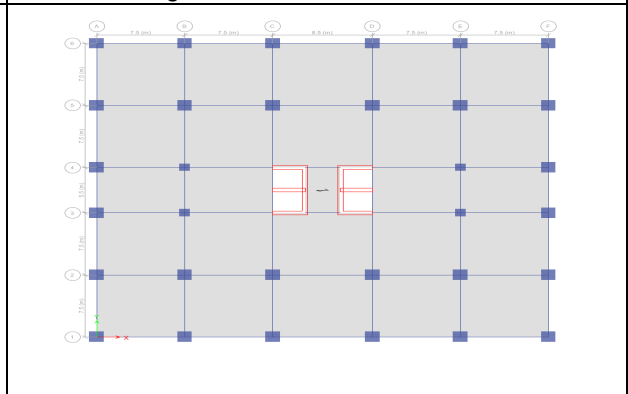
Figure 4. Plan of the Structure IV
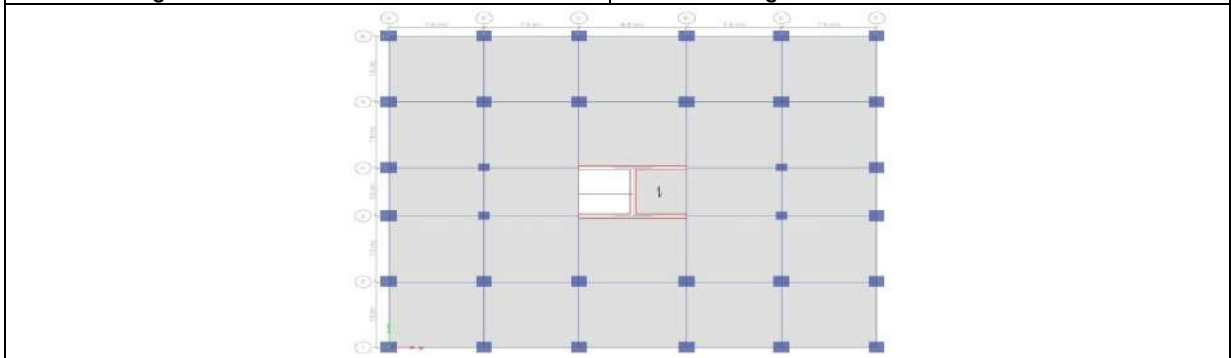
Figure 5. Plan of the Structure V

RESEARCH ARTICLE

Hydrothermal Synthesis of Nickel Vanadate Nanorod for Outstanding Energy Storage Application

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ABSTRACT

To enhance electrochemical performance and to alleviate the energy crisis herein we preferred to develop efficacious vanadium based electrode material. Nickel vanadate nanorod was synthesized by hydrothermal route at different reaction temperature for desired energy storage application. The phase and crystallite nature was analyzed by XRDwhich was found to be 29,32 and 34 nm respectively. The surface morphology of the nanoparticles plays an important role in determining itsproperties. Surface texture of the synthesized nanocomposite scrutinized by SEM analysis which revealed the unique nanorod like structure aligned on a smooth surface. FT-IR spectroscopy was employed to find out the characteristics vibrational modes in the prepared nanostructure. Raman spectroscopy enumerate the vibrational bands of the synthesized nanostructure which is well corrobate with the FT-IR results. The specific capacitance of the synthesized nickel vanadate nanorod was found to be 360 F/g^{-1} at 10 mVs^{-1}.From these aforementioned results it would suggest this could be a hallmark electrode material for future generation capacitors.

Keywords: Nickel vanadate, Hydrothermal, FT-IR, Raman

INTRODUCTION

In this current scenario, the raising of population and demands of fossil fuel are the major crisis in the world-wide. For this instance, the alternative resources like and supercapcitors, batteries and fuel cell are great impact among various researches due to low cost, high efficiency and high power density portable devices. Based up on the crystal size, surface texture, reaction parameters the property of material depends. Among several energy storage devices, supercapacitors have a wide angle because it act as a most prominent candidate due to its durability, high surface area and superior energy density. [1-6]. Supercapacitors are widely depend upon the nature of the material like electric double layer capacitors (EDLC) and fast reversible faradic pseudocapacitors [7-9]. In this era hetero structured hybrid mixed metal oxide nanostructures encountered as a major role in lot of areas chemistry and physics. Due to these consequences everyone focused upon the energy storage applications[10]. Mixed metal oxide has widely encouraging to develop affordable electrode materials,among several mixed metal vanadates like (Cu, Mn, Zn, Co and Ni) nickel shows superior electrical conductiving compare to these electrode materials. Hence there are numerous methods employed for the synthesis of nickel vanadate like sol gel [11],coprecipitaion [12], electrodeposition [13], microwave irradiation [14] and hydrothermal method [15]. On the outset we performed a simple hydrothermal approach of nickel vanadate nanorod for enhancing electrochemical performance. The synthesized nickel vanadate nanorod was evaluated by several physico-chemical methods such as surface texture, phase purity and functional group analysis. Moreover the nickel vanadate nanostructure was explored its potential using three electrode system in supercapacitor domain.

EXPERIMENTAL

Materials
All chemicals were used in this study viz., Nickel (II) nitrate hexahydrate ($Ni(NO_3)_2.6H_2O$), ammonium meta-vanadate (NH_4VO_3), sodium hydroxide (NaOH) Polyvinylidene difluoride (PVDF), *N*-methyl-2-pyrrolidone (NMP) and acetylene black are analytical grade and were purchased from sigma-aldrich. Deionized water was used throughout the study without further purification process.

Synthesis of $Ni_2V_2O_7$ nanorod
In the typical preparation of nickel vanadate was synthesized via hydrothermal process. 2mmol of $Ni (NO_3)_2.6H_2O$ and 1mmol of ammonium metavandate heptahydrate was added into 50 ml of distilled water after that NaOH was added into the solution maintain the pH value at 7. The above mixed solution was maintaining the temperature at 80 °C under magnetic stirring for 30 min. Subsequently, the mixed solution was transferred into 150 ml autoclave, heated at various temperature 160 °C, 180 °C and 200 °C for 10 h. The resulting precipitate was sequentially washed and dried at 80 °C in hot air oven.

RESULTS AND DISCUSSION

XRD Analysis
The structural and phase purity of the prepared samples were further characterized through XRD analysis. Fig.1 shows the XRD spectra of nickel vanadate samples maintained at different temperatures are 160, 180 and 200°C. From the spectra, the diffraction peaks of the samples were well aggregated with the monoclinic $Ni_2V_2O_7$ according to the JCPDS card No 38-0285 [16].The diffraction peaks are $2\theta =(27.2°, 27.8°, 28.9°, 29.8°, 30.9°,32.9°$ and $35.3°)$ corresponds to the (110), (111), (021), (022), (212), (130), (013) planes. When the temperature increases the peak intensities of the samples were gradually increased indicating the well crystalline nature of the samples. The monoclinic structured $Ni_2V_2O_7$ nanoparticles with the similar diffraction peaks were also observed by Ezhil arasi et al and Baba ali et al [17-18]. The average crystallite size of the samples were estimated through Scherer formula in equation 1.

$$D = \frac{k\lambda}{\beta \cos\theta} \qquad (1)$$

Where in, kdenotes the Scherer's constant (0.9), λ refers the wavelength (1.5406 Å), β is the full width half maximum (FWHM) of the line, and θ denotes the diffraction angle. The average crystallite size of the $Ni_2V_2O_7$ maintained at 160, 180 and 200 °C are 29, 32 and 34 nm, When the reaction temperature increases, crystallite size of the samples were gradually increased [19]. Due to the less crystalline size at 160 °C we adopted that temperature for further more characterization techniques.

SEM Analysis
The surface morphology of the prepared samples were characterized through SEM analysis. Fig.2. shows the low and high magnifications SEM images of $Ni_2V_2O_7$. From the Fig.2a, the low magnification SEM image shows the most of the samples are formed as the rod shape with smooth surface with the soft edges. The high magnification image (Fig.2b) of the prepared $Ni_2V_2O_7$ also shows the rod shaped particles with the average size approximately 22 nm. The obtained structure affirms the mobility of electrons will be more easy in supercapacitor applications.

FTIR analysis
The vibrational and chemical composition of the prepared samples was characterized through FTIR analysis. Fig.3. shows the FTIR spectra of $Ni_2V_2O_7$. As from the Fig.2, $Ni_2V_2O_7$ exhibits the two major vibrations in the range of below 1000 cm^{-1}. The small intense peaks aroused at 439 and 654 cm^{-1} are attributed to the stretching vibration of Ni-O modes [20]. And the most intense peak appeared at 816 cm^{-1} is related to the V-O stretching vibrational mode[21]. Additionally, the stretching vibration mode of O-H was appeared in the range of 3469 cm^{-1} and the bending vibration of H-O-H was observed at 1629 cm^{-1}.Hence the synthesized material shows the purity no other impurity peak were observed in the spectrum.

Raman analysis
Raman spectra of prepared $Ni_2V_2O_7$ nanoparticlewas shown in fig. 4. The ten vibrational bands at 143, 192, 315, 413, 539, 591, 709, 787, 857 and 960 cm^{-1}are related to the vibrational modes of $Ni_2V_2O_7$. From the $Ni_2V_2O_7$ spectrum, the peaks at 315, 539 and 591 cm^{-1} are related to the vibrational modes of Ni-O. And the high intense peak at 960 cm^{-1} is assumed to the symmetric stretching vibrational mode of V-O [22]. The peaks at 857 and 787 cm^{-1} are related to the asymmetric stretching vibration of V-O-V and V-O modes. The band assigned at 709 and 413 cm^{-1} is corresponds to the bending vibrational modes of V-O and V=O [23].

Electrochemical analysis
Electrochemical performances were measured to evaluate the potential applications of the $Ni_2V_2O_7$ electrode materials in 6M KOH electrolyte using three electrode system. Fig.5.a. shows the CV curve of prepared $Ni_2V_2O_7$. A quasirectangular shape of the CV curves indicates the capacitive behavior with the range of scan rates from $10\,mVs^{-1}$ to $100\,mVs^{-1}$. The presence of redox species in the CV curve represents the pseudocapacitance behavior of the prepared electrodes. The closely rectangular shape and the redox peaks of the CV was maintained even at the $100\,mV\ s^{-1}$, which may be attributed to the good electrical conductivity of the $Ni_2V_2\ O_7$electrodes. In Fig.5b enumerates the Randles-sevick plot of current versus scan rate curve of $Ni_2V_2O_7$electrodes. The Fig.5 b implies the scan rate increases, anodic current move over to the positive and negative current move over to the negative. increasing the current with respect to the scan rate is due to the increased internal diffusion of resistance which occurs at higher scan rates [24]. Even at high scan rate the redox peaks were still exist, suggesting the good capacitive nature. Further, the specific capacitance of the $Ni_2V_2O_7$ electrode materials were evaluated with the scan rate by use the following Eqn. 2 [25].

$$C_s = s/mk\Delta V \qquad (2)$$

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Where, C_s refers the specific capacitance of the electrode material, m denotes the mass of theelectroactive material (mg), ΔV shows the potential window (V), k refers the scan rate and V is the scan rate (mV s^{-1}) electrodes. The specific capacitance of nickel vanadate are listed in the table1. From the observation nickel enhances the performance of materials due to its superior capacitance rate. As shown in Table 1, electrode achieved a highest specific areal capacitance at 10 mVs^{-1}scan rate. It corroborates the good electrochemical rapport Ni2V2O7and provides a good conductive route for the rapid ion transportation [26]. Fig. 5a demonstrates that the specific capacitance of each electrode decreased with an increase of scan rate. The declination of specific capacitance is due to time limitation for insertion and exertion of the electrolyte ion from the electrode [27].

CONCLUSION

Nickel vanadate nanorod were synthesized by a cost-effective hydrothermal approach at different temperature and owing to the lesser crystalline size the reaction temperature fixed for furthermore characterization techniques. XRD pattern portrays the existence of monoclinic phase and corroborate with the JCPDS card number, no other impurity peak were observed here and the crystallite size found to be 29, 32 and 34 nm. Surface texture revealed the distinct nanorod was uniformly distributed over the smooth surface. The several functional groups related to nickel vanadate nanorods such as Ni-O and V-O vibrational modes had confirmed by the FT-IR and Raman spectroscopy. The specific capacitance of the nickel vanadate exhibited as 360 F/g^{-1} at 10 mVs^{-1}manifest pseudocapacitance nature. These research findings will be a new avenue for developing nickel based future energy storage devices.

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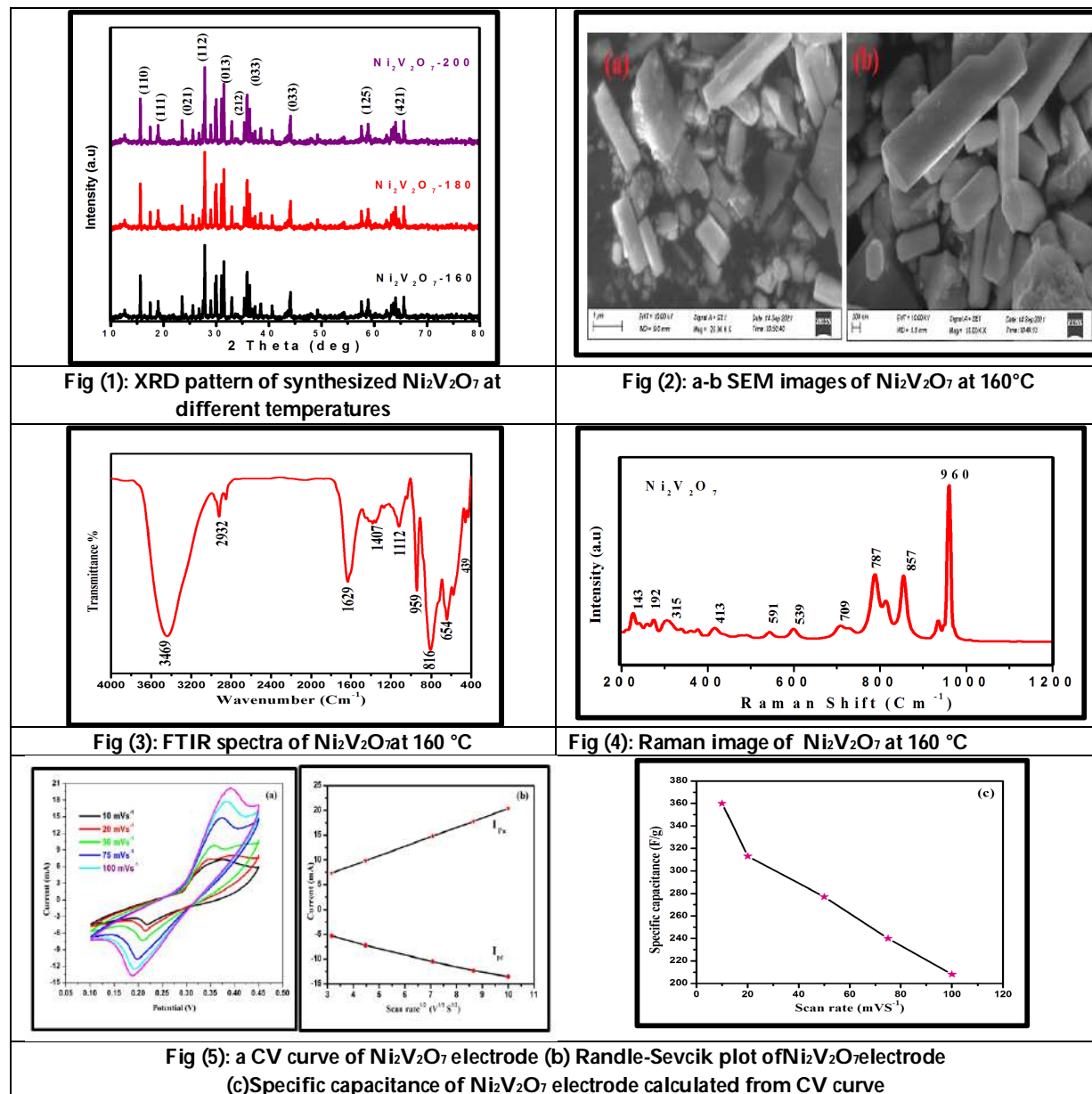
Table 1: Specific capacitance values for $Ni_2V_2O_7$ electrode enumerated from CV curve

Scan rate mV s^{-1}	$Ni_2V_2O_7$ Fg^{-1}
10	360
20	313
50	277
75	240
100	208

Fig (1): XRD pattern of synthesized Ni₂V₂O₇ at different temperatures

Fig (2): a-b SEM images of Ni₂V₂O₇ at 160°C

Fig (3): FTIR spectra of Ni₂V₂O₇at 160 °C

Fig (4): Raman image of Ni₂V₂O₇ at 160 °C

**Fig (5): a CV curve of Ni₂V₂O₇ electrode (b) Randle-Sevcik plot ofNi₂V₂O₇electrode
(c)Specific capacitance of Ni₂V₂O₇ electrode calculated from CV curve**

RESEARCH ARTICLE

L^α − Fuzzy Almost Subgroups of a Group G

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ABSTRACT

In this paper, the idea of L^α-fuzzy subset,L^α-fuzzy almost subset of a group is described and its properties are determined. Thought this paper, fuzzy sub groups of L^α-Fuzzy sets have been well studied in the case where the lattice is not distributive. The aim of this current paper is to define the concept of L^α-Fuzzy Almost Subgroup (L^α-FASG) and investigate some of its properties.

Keywords - L^α-fuzzy subset,L^α-fuzzy almost subset and L^α-Fuzzy Almost Subgroup

INTRODUCTION

Zadeh [19] introduced the concept of fuzzy sets in 1965. In [13] Rosenfeld utilized this idea to foster the hypothesis of fuzzy groups. Truth be told, numerous fundamental properties in bunch hypothesis are viewed as extended to fuzzy groups. The theory of L- fuzzy sets was initiated by Goguen [8]. In [18] Wang-Jin Liu studied fuzzy subgroups and fuzzy ideals in 1982. In [15] Sivaramakrishna Das researched Fuzzy Groups and Level Sub groups.In [1] Ajmal defined Homomorphism of groups and proved some correspondence theorem and fuzzy quotient groups in1994. Anthony and Sherwood [2,3] formulated the definition of fuzzy subgroup based on t-norm. In [4,7] Bhakat and Das have studied the concept of fuzzy subgroups. In [5] Bhattachar and Mukherjee have studied the concept of fuzzy groups. Chakrabatty and Khare [6] have presented the thought of fuzzy homomorphism between two groups and concentrated on its impact to the fuzzy sub groups. In [14] Sharma formulated the definition of α -fuzzy subgroup to be fuzzy subgroup and discussed some properties of α-Fuzzy subgroups. In [16,17] Veerammal and Velammal have defined the concept of L - Fuzzy Almost Ideals and L-Fuzzy Almost Subgroup (LFASG) and investigate some of its properties. The point of this current paper is to characterize the L^α-FASG which is a speculation of L- fuzzy subgroup and research a portion of its properties.

Preliminaries

Let X be a nonempty subset, $(L, \leq, \vee, \wedge)$ be a complete distributive lattice, which has least and topmost elements, say 0 and 1 respectively. Applicable and necessary definitions are recalled in this section.

Definition 2.1 [17] Let G be a group with the identity element e. Let L be a lattice $(L, \leq, \vee, \wedge)$ not necessarily distributive with least and most prominent component 0 and 1 separately. Then L-fuzzy subset H defined by $\mu: G \rightarrow L$ with μ (e)=1 is said to be L-fuzzy almost subgroup (LFASG) if,

(i) $\mu(xy) \not< \mu(x) \wedge \mu(y)$, for all $x, y \in G$

(ii) $\mu(x^{-1}) \not< \mu(x)$, for all $x \in G$

Definition 2.2 [17] If A is a L-fuzzy set of X defined by $\mu: X \rightarrow L$ then the Almost level set $\approx A_t$ is defined as $\approx A_t = \{x \in X | \mu(x) \not< t\}$.

Proposition 2.3 [17] If $t \leq s$, $\approx A_t \supseteq \approx A_s$.

Definition 2.4 [14] Let $A: G \rightarrow [0,1]$ be a fuzzy subset of a group G. Let $\alpha \in [0,1]$. Then the fuzzy set A^α of G is called the α-fuzzy subset of G and is defined as $A^\alpha(x) = \min\{A(x), \alpha\}$, for all $x \in G$.

Definition 2.5 Let $A: G \rightarrow [0,1]$ be a fuzzy subset of a group G. Let $\alpha \in [0,1]$. Then A is called the α-fuzzy subgroup of G if

(i)　　　$A^\alpha(xy) \geq \min\{A^\alpha(x), A^\alpha(y)\}$, for all $x, y \in G$

(ii)　　$A^\alpha(x^{-1}) \geq A^\alpha(x)$, for all $x \in G$.

Definition 2.6 [1,6] Let f be a homomorphism of group G_1 into a group G_2. Let A and B are the two fuzzy subsets of G_1 and G_2 respectively. Then f(A) and $f^{-1}(B)$ are respectively the image of fuzzy set A and the inverse image of fuzzy set B, defined as

$$f(A)(y) = \begin{cases} \sup\{A(x) / x \in f^{-1}(y)\} & if f^{-1}(y) \neq \emptyset \\ 1 & if f^{-1}(y) = \emptyset \end{cases}, \text{ for every } y \in G_2 \text{ and } f^{-1}(B)(x) = B(f(x)), \text{ for every } y \in G_1$$

L^α-Fuzzy Subset

In this part, the idea of L^α-fuzzy subset of a g group is characterized and its properties are determined.

Definition 3.1 Let $\mu_{al}: G \rightarrow L$ be a L-fuzzy subset of a group G. Let $\alpha \in L$. Then the α-Lattice fuzzy subset of G ($L^\alpha - \alpha$ Lattice), denoted by $\mu_{al}{}^\alpha$ is known as the L^α-fuzzy subset of G and is characterized by $\mu_{al}{}^\alpha(x) = \mu_{al}(x) \wedge \alpha$, $\forall x \in G$.

Definition 3.2 If A will be a α-Lattice fuzzy set of X characterized by $\mu_{al}: X \rightarrow L$ then the α-Almost level set $\approx \mu_{al}{}^\alpha$ is characterized as $\approx \mu_{al}{}^\alpha_t = \{x \in X | \mu_{al}{}^\alpha(x) \not< t\}$.

Definition 3.3 A homomorphism of group G_1 into G_2 be $\mu_{al}: G_1 \rightarrow G_2$. Let $\mu_{al_A}: G \rightarrow L$ and $\mu_{al_B}: G \rightarrow L$ be two L-fuzzy subsets of a group G_1 and G_2 respectively. Then $f(\mu_{al_A})$ and $f^{-1}(\mu_{al_B})$ are respectively the image of L-fuzzy set μ_{al_A} and the inverse image of fuzzy set μ_{al_B}, defined as

$$f(\mu_{al_A})(y) = \begin{cases} \vee\{\mu_{al_A}(x): x \in f^{-1}(y)\} & if f^{-1}(y) \neq \emptyset \\ 1 & if f^{-1}(y) = \emptyset \end{cases}, \text{ for every } y \in G_2 \text{ and}$$

$f^{-1}(\mu_{al_B})(x) = \mu_{al_B}(f(x))$, for every $y \in G_1$.

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Example 3.4 If $\mu_{al}(x) = \begin{cases} e \, if \, x = 1 \\ a \, if \, x = 0.3 \\ b \, if \, x = 0.7 \\ c \, if \, x = 0.5 \\ 0 \, if \, if \, x = 0 \end{cases}$. Take $\alpha = 0.25$ then $\mu_{al}{}^\alpha(x) = \begin{cases} 1 \, if \, x = e \\ 0.7 \, if \, x = a \\ 0.9 \, if \, x = b \\ 0.8 \, if \, x = c \end{cases}$

Let L be a lattice $(L, \le, \vee, \wedge)$. Defined $\mu_{al}: G \rightarrow L$ and $\mu_{al}{}^\alpha:G \rightarrow L$ then μ_{al} and $\mu_{al}{}^\alpha$ are represented by the following Hasse diagrams (**Figure 3.1** and **Figure 3.2**).

In the accompanying **Table 3.1**, shows that the confirmation of $\mu_{al}{}^\alpha(x)$ as follows:

Theorem 3.Theorem 3.5 Let μ_{al_A}:G→L and μ_{al_B}:G→ L be two L^α- fuzzy subsets of a group G. Then $\mu_{al_{A \cap B}}{}^\alpha$ = $\mu_{al_A}{}^\alpha \cap \mu_{al_B}{}^\alpha$

Theorem 3.6 Let f be a mapping from X to Y and A and B be two L^α- fuzzy subsets of X and Y respectively, then (i) $f(\mu_{al_A}^\alpha) = (f(\mu_{al_A})^\alpha)$

(ii) $f^{-1}(\mu_{al_B}^\alpha) = (f^{-1}(\mu_{al_B}))^\alpha$

L^α −Fuzzy Almost Subgroups

In this segment, the definition of L^α-fuzzy subset, L^α-fuzzy almost subset of a group G have been presented . Furthermore, the instances of L^α-fuzzy subset, L^α-fuzzy almost subset of a group G have been examined exhaustively and furthermore some corresponding theorems are demonstrated on it.

Definition 4.1 Let G be a group with the identity element e. Let L be a lattice (L, $\le$, $\vee$, $\wedge$) not necessarily distributive with least and most prominent component 0 and 1 separately. Then, at that point, L^α-fuzzy subset of G defined by $\mu_{al}: G \rightarrow L$ with $\mu_{al}(e)=1$ is supposed to be L^α- fuzzy almost subset if, $\mu_{al}{}^\alpha(x) \not< \mu_{al}(x) \wedge \alpha$ (or) $\mu_{al}{}^\alpha(x) \not> \mu_{al}(x) \wedge \alpha$, $\forall x \in$ G.

Definition 4.2 Let $\mu_{al}: G \rightarrow L$ be a L- fuzzy subset of a group G. Let $\alpha \in L$ .Then the L-fuzzy set $\mu_{al}{}^\alpha$ of G is called the L^α-fuzzy almost subgroup (L^α-FASG) of G if
- $\mu_{al}{}^\alpha(xy) \not< \mu_{al}{}^\alpha(x) \wedge \mu_{al}{}^\alpha(y)$, $\forall x, y \in$ G.
- $\mu_{al}{}^\alpha(x^{-1}) \not< \mu_{al}{}^\alpha(x)$, $\forall x \in$ G.

Proposition 4.3 A function $\mu_{al}: G \rightarrow L$ is a L-FASG iff $\mu_{al}(xy^{-1}) \not< \mu_{al}(x) \wedge \mu_{al}(x)$,$\forall x, y \in$ G

Proposition 4.4 A function $\mu_{al}: G \rightarrow L$ is a L-FASG then
 (i) $\mu_{al}(x) \not> \mu_{al}(e)$,$x \in$ G where e is the identity element of G
(ii) $\mu_{al}(xy^{-1}) = \mu_{al}(e) \Longrightarrow \mu_{al}(x) = \mu_{al}(y)$,$\forall x, y \in$ G

Example 4.5 Let G = $\{e, a, b, c\}$, where $a^2 = b^2 = e$ and $ab = ba$ be the Klein-4 group, whose Composition Table 4.1 is as follows:

Case-I Let L be a lattice (L, $\le, \vee, \wedge$)defined characterized by the accompanying Hasse chart the Figure 4.1. Define $\mu_{al}: G \rightarrow L$

If $\mu_{al}(x) = \begin{cases} e\, if\, x = 1 \\ b, c\, if\, x = 0.7 \\ a\ \ if\, x = 0 \end{cases}$ then, at that point, it very well may be checked that μ_{al} characterizes a LFASG.

From Table 4.3 and Table 4.4, we ge, μ_{al} is a **L-FASG.**

Case-II Let L be a lattice $(L\, , \leq, \vee, \wedge)$ defined by the following Hasse diagram (Figure 4.2)
Consider $\mu_{al}{}^{\alpha}: G \longrightarrow L$. Take $\alpha = 0.65$.

If $\mu_{al}(x)^{\alpha} = \begin{cases} 1\, if\, x = e \\ 0.8\, if\, x = b, c \\ 0\ \ if\, x = a \end{cases}$

then, at that point, it tends to be checked that μ_{al} characterizes a $L^{\alpha} - FASG$.

From Table 4.6 and Table 4.7, we ge, μ_{al} is a $\boldsymbol{L^{\alpha} - FASG}$

Theorem 4.6 A function $\mu_{al}: G \longrightarrow L$ be a L^{α}- FASG then
(i) $\mu_{al}{}^{\alpha}(x) \not\succ \mu_{al}{}^{\alpha}(e)$, for all $x \in$ G, where e is the identity element of G.
(ii) $\mu_{al}{}^{\alpha}(xy^{-1}) = \mu_{al}{}^{\alpha}(x) \Rightarrow \mu_{al}{}^{\alpha}(x) = \mu_{al}{}^{\alpha}(y)$, $\forall\, x, y \in$ G.

Theorem 4.7 A function $\mu_{al}: G \longrightarrow L$ be a L-FASG then μ_{al} is also L^{α}- FASG of G.

Remark 4.8 The opposite of the above hypothesis is need not be valid.

Theorem 4.9 A function $\mu_{al}: G \longrightarrow L$ be a L-fuzzy subset of a group G such that $\mu_{al}(x^{-1}) \geq \mu_{al}(x)$, $\forall\, x \in$ G. Let $\alpha \leq p$, where $p = \wedge\{\mu_{al}(x) / x \in G\}$. Then μ_{al} is a L^{α}-FASG of G.

Theorem 4.10 Intersection of any two L^{α}-FASG of a group G is also an L^{α}-FASG of a group G.

CONCLUSIONS

In this paper we have fostered the idea of L-fuzzy almost subgroup. We likewise examine the impact of homomorphism on L-fuzzy almost subgroup. We have demonstrated some theorems on almost level set and L-fuzzy almost subgroup.

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Table 4.1

$*$	e	a	b	c
e	e	a	b	c
a	a	e	c	b
b	b	c	e	a
c	c	b	a	e

Table 4.2 Multiplication table of the group

$*$	1	0	0.7	0.7
1	1	0	0.7	0.7
0	0	1	0.7	0.7
0.7	0.7	0.7	1	0
0.7	0.7	0	0	1

Table 4.3- Axiom (i) verification for L-FASG

x \ y		1	0	0.7	0.7	Conclusion
1	$\mu_{al}(x) \wedge \mu_{al}(y)$	e	a	b	c	Satisfied
	$\mu_{al}(xy)$	e	a	b	c	
0	$\mu_{al}(x) \wedge \mu_{al}(y)$	a	a	a	a	satisfied
	$\mu_{al}(xy)$	a	e	c	b	
0.7	$\mu_{al}(x) \wedge \mu_{al}(y)$	b	a	b	a	Satisfied
	$\mu_{al}(xy)$	b	c	e	a	
0.7	$\mu_{al}(x) \wedge \mu_{al}(y)$	a	a	a	a	satisfied
	$\mu_{al}(xy)$	c	b	a	e	

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Table 4.4 - Axiom (ii) verification for L-FASG

x	$\mu_{al}(x)$	$\mu_{al}(x^{-1})$	conclusion
1	e	e	verified
0	a	a	verified
0.7	b	b	verified
0.7	c	c	verified

Table 4.5- Multiplication table of the group

$*$	e	a	b	c
e	1	0	0.7	0.7
a	0	1	0.7	0.7
b	0.7	0.7	1	0
c	0.7	0.7	0	1

Table 4.6- Axiom (i) verification for $L^{\alpha} - $ FASG

x \ y		e	a	b	c	Conclusion
e	$\mu_{al}(x)^{\alpha} \wedge \mu_{al}(y)^{\alpha}$	1	0	0.8	0.8	Verified
	$\mu_{al}(xy)^{\alpha}$	1	0	0.8	0.8	
a	$\mu_{al}(x)^{\alpha} \wedge \mu_{al}(y)^{\alpha}$	0	0	0	0.8	verified
	$\mu_{al}(xy)^{\alpha}$	0	1	0.8	0.8	
b	$\mu_{al}(x)^{\alpha} \wedge \mu_{al}(y)^{\alpha}$	0.8	0	0.8	0	Verified
	$\mu_{al}(xy)^{\alpha}$	0.8	0.8	1	0	
c	$\mu_{al}(x)^{\alpha} \wedge \mu_{al}(y)^{\alpha}$	0.8	0	0	0.8	Verified
	$\mu_{al}(xy)^{\alpha}$	0.8	0.8	0	1	

Table 4.7 -Axiom (ii) verification for $L^{\alpha} - $ FASG

x	$\mu(x)$	$\mu(x^{-1})$
e	1	1
a	0	0
b	0.8	0.8
c	0.8	0.8

| **Figure 3.1. Hasse chart** | **Figure 3.2 Hasse diagrams** | *Figure 4.1* **Hasse chart** | *Figure 4.2* **Hasse diagram** |

RESEARCH ARTICLE
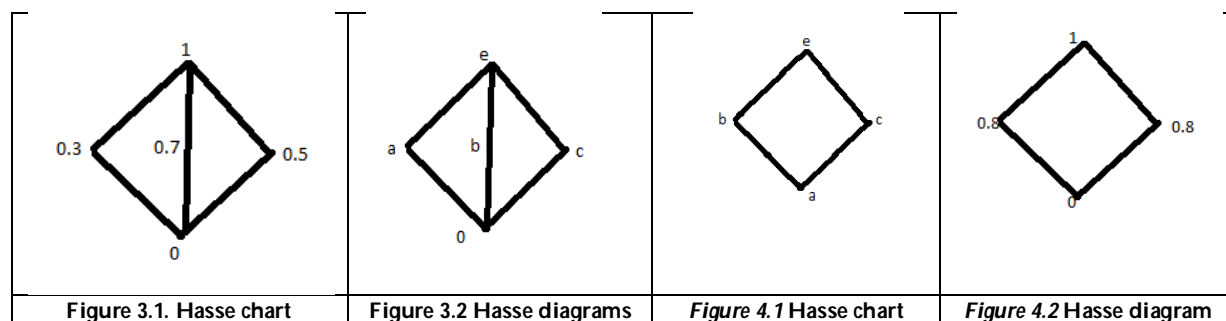
Response of Integrated Nutrient Management on Sugarcane Yield and Ratoon Yield

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ABSTRACT

Field experiment were conducted in sandy loam soil at Mamangalam village, Kattumannarkoil taluk, Cuddalore district, Tamil Nadu. The soils of Mamangalam was classified as *Typic ustipsamments* having sandy texture, the available nutrient status was low in N, medium in P and low in K. In plant crop experiment treatment consisted of main plot treatments Control, FYM @ 25 t ha-1, Seasoned Pressmud @ 25tha-1 and Biocompost @ 5 t ha-1. The sub plot treatments were 100% RDF (N: P_2O_5:K_2O @ 275:62.5: 112.5 kg ha-1), 75% RDF + Lignite flyash @ 25 t ha-1, 75% RDF + Humic acid @ 50 kg ha-1, 75% RDF + Lignite fly ash @ 25 t ha-1 + Humic acid @ 50 kg ha-1. The design followed was split plot design. The treatments were replicated thrice. The result of the plant crop experiment conducted in sandy soil indicated that the influence of main plot and sub plot treatments were significant. Among main plot treatments application of Seasoned Pressmud @ 25 t ha-1 recorded highest cane weight of 1.27 kg millable cane population of 123.9 (x1000' ha-1) plant cane yield of 153.5 t ha-1 and ratoon cane yield of 132.9 t ha-1. With regard to sub plot treatments, plot receiving 75% RDF + Lignite Fly ash @ 25 t ha-1 + Humic acid @ 50 kg ha-1 registered maximum cane weight of 1.24 kg, millable cane population of 119.8 (X1000' ha-1) and ratoon cane yield of 132.7 t ha-1.

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INTRODUCTION

India is a second largest producer of sugarcane cultivating in 4.9 million ha of land and the production of 376.905 million tonnes of cane per year. The productivity is 77.6 t ha^{-1} (Sugarcane Statistics, 2020). The country's requirement by 2025 AD has been projected at 625 Mt which mean that there is need to raise the productivity of sugarcane and sustain the same (Sundara, 1998). Integrated nutrient management (INM) involves the integrated use of mineral fertilizers together with organic manures/ industrial agricultural wastes in suitable combination complementing each other to optimize input use and maximize production and sustain to same without impairing the crop quality of soil health. It enables gainful utilization of waste or under utilized renewable resources. The present study was designed to find out the effect of soil and nutrient management practices involving organics/industrial by-products and fertilizers on yield-attributes and yield of sugarcane.

MATERIALS AND METHODS

The field experiment was conducted in farmer's field in Mamangalam village (Sandy loam) of Chidambaram Taluk, Cuddalore district, Tamilnadu. Soil properties are given in Table 1. The experiment was laid out in a split plot design. The main plot treatments included M_1 – Control; M_2 FYM @ 25 t ha^{-1}; M_3 – Seasoned Pressmud @ 25 t ha^{-1} and M_4 – Biocompost BC @ 5 t ha^{-1}, Sub plot treatments constituted S_1-100% recommended dose of fertilizer (RDF) (N, P, K @ 275:62.5: 112.5 kg ha^{-1}), S_2 – 75% RDF + Lignite flyash LFA @ 25 t ha^{-1}, S_3 – 75% RDF + Humic acid (HA) 50 kg ha^{-1}, S_4 - 75% RDF + 25 t LFA ha^{-1} + 50 kg HA ha^{-1}. The chemical composition of SPM, FYM, BC, LFA and HA materials is given in Table 2. The yield attributes include number of millable canes at harvest ($\times$ 1000 ha^{-1}) and weight of millable cane (kg). Soil samples were shade-dried, powdered and sieved through 2 mm sieve were used for analysis by following the standard procedures. After the harvest main crop in the same experimental layout, sugarcane crops were allowed to grow as ratoon crop to study the residual effect of treatments. The individual plots in case of ratoon crops received only recommended dose of N, P_2O and K_2O fertilizers. The crops were grown following the recommended package of practices and harvested at maturity. The cane harvested from each experiment was weighed and expressed as tonnes per hectare (t ha^{-1}).

Individual cane weight and number of millable cane
The data on individual cane weight (kg) and number of millable cane population ($\times$ 1000 ha^{-1}) recorded at harvest were presented in Table 3 showed that the organic manure treatments had significant influence on individual cane weight and number of millable cane. Treatment M_3 seasoned pressmud @ 25 t ha^{-1} recorded maximum individual cane weight of 1.27 kg and millable cane population of 123.9 ($\times$ 1000 ha^{-1}) in sandy loam soil. In sugar cane production individual cane weight and number of millable canes assure practical significance as they are directly related to productivity. It has been established that an increase in tillering ability of sugarcane plant leads to increase in the number of millable canes which is one of the important yield attributes responsible for boosting the productivity of sugarcane (Babu, 2004). The advantage of organic manures is quite obvious, as these provided a steady supply of nutrients leading better growth of plants. Moreover, the increased availability of P and K in addition to other plant nutrients released by the organic manures might have contributed in enhancing yield-attributes. The positive impact of availability of individual plant nutrients and humic substances from manure and balanced supplement of nitrogen through inorganic fertilizers might have induced cell division expansion of cell wall, meristematic activity, photosynthetic efficiency and regulation of water intake into the cells, resulting in the enhancement of yield parameters (Jha *et al.*, 2019). The increase in yield characters of sugarcane due to integrated application of pressmud and inorganic fertilizers might be attributed to the reduced tiller mortality resulting in more number of millable cane. Pressmud itself contain 21 per cent organic carbon along with other macro and micronutrients which promote the yield. The differences in individual cane weight were significant among industrial by-product treatments. Among

the sub plot treatments comprising of LFA @ 25 t ha^{-1} + HA @ 50 kg ha^{-1} (S$_4$) registered maximum individual cane weight of 1.24 kg and maximum population of 119.8 ($\times$1000 ha^{-1}) in sandy loam soil. The addition of LFA and HA exhibited superiority by registering significantly higher millable cane and individual cane weight in experimental field. The Flyash acts as a soil increases the yield attributes (Das *et al.,* 2013). Humic acid influences the growth of higher plants favourable through increase in cane length (Amir Rahimi *et al.,* 2020).

Plant cane yield

Among organic manure treatments, M$_3$ (SPM @ 25 t ha^{-1}) recorded maximum cane yield of 153.50 t ha^{-1} in sandy loam soil (Table 4). The increase in sugarcane yield due to application of organic manures can be attributed to the increased availability of all major nutrients into soil. Application of pressmud along with inorganic fertilizer, followed by FYM and inorganic fertilizers resulted in higher cane yield. In sugarcane cultivation, the cane yield is the ultimate product that decides the benefit accrued out of it. The higher response of sugarcane due to application of organic manures may be attributed to the availability of plant nutrients by the solubilizing effect of the decomposing manures with steady release of plant nutrients over longer periods. The increased soil organic carbon content and consequently improved soil physico-chemical properties with application of organic manures might have contributed to the increased cane yield. Application of manures such as SPM, FYM and BC might have improved to physical condition of soil by reducing bulk density and increasing soil macrospore for better root proliferation and finally reflected on cane yield (Bokhtiar *et al.,* 2015). The appreciable increase in cane yield due to addition of SPM is attributed to improvement in various growth and yield attributes such as weight and number of millable cane at harvest (Srivastava *et al.* 2006). Industrial by-products exhibited significant difference on cane yield. Treatment.

S$_4$ (LFA @ 25 t ha^{-1} + HA @ 50 kg ha^{-1}) recorded maximum cane yield of 143.37 t ha^{-1}. The flyash application had greater effect on improving cane yield (Rajinderkaur and Dinesh Goyal, 2014). Beneficial role of humic acid in improving sugarcane productivity had been highlighted by Rajula Shanthy *et al.* (2014).

Ratoon cane yield

Application of SPM along with inorganic fertilizers followed by FYM and fertilizers resulted in the highest cane yield in ratoon crop (Table 4). Among organic manure treatments, treatment M$_3$ (SPM @ 25 t ha^{-1}) proved superior to control by recording 32.02% higher yield (144.2 t ha^{-1}) in sandy loam soil. Application of the organic manures application decreased the bulk density, salt accumulation and increased the *in situ* saturated hydraulic conductivity due to increased soil organic carbon content. The higher response of ratoon was due to residual effect of added organic manures such as SPM, FYM, BC on plant crop and would have improved the soil physico-chemical and biological properties to some extent (Ramalingaswamy *et al.* 1995). Among sub-plot treatments the LFA @ 25 t ha^{-1} + HA @ 50 kg ha^{-1} (S$_4$) proved superior by registering an increased ratoon yield (132.7 t ha^{-1}) of 10.7% over control. The LFA might have served as a good amendment and source of nutrients even after the plant crop (Kumari Manimuthuveeral, 2014).

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Table 1. Initial soil properties of the experimental field

Properties	Value
Coarse sand (%)	49
Fine sand (%)	40
Silt (%)	5
Clay (%)	5
Textural class	Sandy
Taxonomical classification	*Typic ustipsamments*
pH	7.12
EC (dS m^{-1})	0.24
Organic carbon (g kg^{-1})	5.45
CEC (c. mol (p+) kg^{-1})	17.1
KMnO$_4$-N (kg ha^{-1})	243.0
Olsen-P (kg ha^{-1})	22.5
NH$_4$OAC-K (kg ha^{-1})	105.2

Table 2. NPK content of materials

Materials	N	P	K
SPM (%)	1.26	3.83	1.46
FYM (%)	0.79	0.92	0.80
BC (%)	2.0	2.0	3.0
LFA (mg kg^{-1})	0.28	–	43
HA (%)	0.38	0.007	7.72

Table 3. Effect of organic manures and industrial by-products on number of individual cane weight (kg) and no. of millable canes (x000 ha^{-1}) at harvest

Main plot	Individual cane weight (kg)					No. of millable cane (X 1000 ha^{-1})				
Sub plot	Control (M$_1$)	FYM (M$_2$)	SPM (M$_3$)	BC (M$_4$)	Mean	Control (M$_1$)	FYM (M$_2$)	SPM (M$_3$)	BC (M$_4$)	Mean
RDF (S$_1$)	1.12	1.17	1.22	1.16	1.17	105.2	112.5	120.0	117.7	112.3
RDF+LFA (S$_2$)	1.15	1.26	1.30	1.21	1.23	109.4	123.8	126.4	117.1	119.1
RDF+HA (S$_3$)	1.13	1.17	1.23	1.17	1.17	105.8	113.3	121.7	111.9	114.5
RDF+LFA + HA (S$_4$)	1.16	1.28	1.33	1.22	1.24	110.1	124.6	127.5	117.1	119.8
Mean	1.15	1.22	1.27	1.18	1.20	107.6	118.6	123.9	115.7	116.4

	S.Ed.	CD (p=0.05)	S.Ed.	C.D. (p=0.05)		S.Ed.	CD (p=0.05)	S.Ed.	C.D. (p=0.05)
M	0.01	0.04	0.01	0.04		1.65	4.05	1.76	4.31
S	0.03	0.06	0.02	0.05		2.70	5.57	2.77	5.73
M x S	0.05	NS	0.05	NS		4.96	NS	5.12	NS
S x M	0.06	NS	0.05	NS		5.40	Ns	5.55	NS

Table 4. Effect of organic manures and industrial by-products on plant and ratoon yield of sugarcane (t ha^{-1})

Main plot	Plant cane yield (t ha^{-1})					Ratoon cane yield (t ha^{-1})				
Sub plot	Control (M$_1$)	FYM (M$_2$)	SPM (M$_3$)	BC (M$_4$)	Mean	Control (M$_1$)	FYM (M$_2$)	SPM (M$_3$)	BC (M$_4$)	Mean
RDF (S$_1$)	117.0	132.1	146.47	127.10	130.4[b]	105.2	124.0	136.2	113.9	119.8[b]
RDF+LFA (S$_2$)	120.6	149.0	157.40	135.20	140.55[a]	111.4	139.8	149.2	119.5	130.0[a]
RDF+HA (S$_3$)	119.0	134.0	148.20	130.00	133.33[b]	107.4	125.0	137.6	116.6	121.6[b]
RDF+LFA+HA (S$_4$)	122.4	152.4	162.30	136.35	143.37[a]	113.0	142.6	153.9	121.2	132.7[a]
Mean	119.7[d]	140.9[b]	153.50[a]	133.16[c]	136.84	109.2[c]	132.9[ab]	144.2[a]	117.8[bc]	126.0

	S.Ed.	CD (P=0.05)	S.Ed.	CD (p=0.05)
M	2.52	6.17	2.62	6.42
S	3.58	7.39	3.50	7.24
M×S	NS	NS	6.62	NS
S×M	NS	NS	7.01	NS

SHORT COMMUNICATION ARTICLE

Exposure to Non-Ionizing Electromagnetic Radiation-Human Health

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ABSTRACT

Electromagnetic waves are due to the intermittent vibrations between electric and magnetic fields. Environmental submission to unnatural electromagnetic fields has widened as new advances have linked an increasing number of sources of reproducing them. The article presents the case of appliances like Mobile Phones that emit Non-Ionizing Radiation (NIR) and portrays the threat due to electromagnetic radiation. Though globally it has become mandatory to restrict SAR (Specific Absorption Rate) values w.r.t. gadgets generating electromagnetic radiation, still some precautions need to be followed to limit the dangers arising from NIR. Because of its unavoidable nature, wide applications, adverse effects, some health risk tests are essential. The article also suggests a couple of careful steps that can reduce the danger of these risks.

Keywords: Electromagnetic radiation, Mobile phone, Non-Ionizing Radiation (NIR), SAR.

INTRODUCTION

The current electromagnetic framework is continually being modified by humans due to the rapid increase in innovative events and these electromagnetic waves provide unlimited benefits as well as carry probable risks through uncontrolled and extreme radiation outputs. The article presents the case of gadgets that emit electromagnetic radiation, clarifies the health risks and also presents some precautionary estimates that can lessen the danger of these risks from electromagnetic radiation. Innovative advances with adjustments in social behavior have led to an escalation of exposure to artificial electromagnetic fields. Surely all of us, sooner or later, are introduced to electric fields, both at home and at work, with electrical power, residential machines, mechanical hardware and information transmission.

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What are electromagnetic waves (EM waves)?

EM waves arise due to the motion of charged particles. EM radiation is a form of energy transmitted by means of charged particles. They travel in space, in air and in other subjects. Low-frequency EM waves are called electromagnetic fields, and higher frequencies are called electromagnetic radiation

Classification of Electromagnetic (EM) radiation

EM radiation is classified into ionizing and Non-Ionizing Radiation (NIR) depending on their energy and frequency. Ionizing radiation is High-Frequency EM waves like X-rays & γ- rays. They hold adequate energy to create ionization by breaking the bonds that hold atoms together. The portion in the electromagnetic field that doesn't have enough energy to break bonds is commonly called non-ionizing radiation (ex: UV, infrared, radio frequency radiation and microwave fields). NIR cannot cause ionization, but it appears to have other natural effects, such as by heating or inducing electric currents in tissues [1]. Direct amounts of NIR can damage fabrics with heat. This mainly concerns people who work with large sources of NIR equipment and tools [2]. There are 4- subdivisions of EM radiation w.r.t. frequency.

Rationales of EM fields & EM waves

While electric fields are related to the existence of charges, magnetic fields on the other hand are due to the physical movement of the charges. Electric field (E) which is expressed in (V / m) volts per meter can exert a force on a charge. Subsequently, magnetic fields also can exert force on moving charges.The Magnetic field is indicated by "B"– the Magnetic flux density (units: webers/m^2 or Tesla), or as, "H"-the Magnetic field strength, (units: A/m).

"B" & "H" are associated as: B = μH (where "μ" is magnetic permeability)

Radio frequencies (or Radio waves) are a kind of electromagnetic radiation which has electrical and magnetic components that go together in a similar direction, but in regions perpendicular to each other. The region where the waves materialize is called Electromagnetic Field (EMF) [3] and the waves are produced due to motion of electric charges on conductive metal objects (ex: transmitting antennas). Every radiation has specific frequency and wavelength. Wavelength is the distance between 2successive points (crests or troughs) in the adjacent cycles of a waveform (signal) and generally identified in meters (m) or centimeters (cm). In case of radiation like infrared, visible light, ultraviolet, the wavelength is stated in units of angstroms (1Å = 10^{-10} m). Total number of waves that pass a fixed point within a specified period of time is frequency and measured in units of hertz (Hz).In the case of energy, EM wave energy is directly proportional to its frequency and inversely proportional to its wavelength.RF (radio frequency) energy is given in watts per kilogram (W / kg) [3].Electromagnetic radiation is emitted by many devices that people often encounter. Microwave ovens, cell phones, radars, industrial heaters, TV screens, refrigerators, etc. are few devices that people use regularly.

Properties of electromagnetic waves (EM waves)

Electromagnetic waves are mass less but carry energy and do not need any means to propagate. They possess momentum and they also can exert pressure (known as radiation pressure). According to Max Planck, EM wave's energy is directly proportional to frequency of the wave (E = hf). Wavelength (λ) and frequency (f) of the wave are associated through the speed of light by the relation: c = $f\lambda$.EM waves are separated into different types on the basis of frequency of various regions of the spectrum. Humans can see electromagnetic waves whose frequency is between 4.3×10^{14} Hz and 7.5×10^{14} Hz (or of wavelength from 380 nm to 700 nm). Example: visible light ranging from purple to red. Light (which belongs to the visible region) is a tiny part of the spectrum. EM waves with higher energy than visible light include UV light, X-rays and γ- rays [3]. Infrared light, μ-waves, radio waves are called low energy waves.

Energy of an EM wave

The energy inside an EM wave is connected with electric (E) and magnetic (B) fields. Energy density (energy per unit volume) for an Electric Field is: $\frac{1}{2}\varepsilon_0 E^2$ (ε_0 is permittivity & E is electric field intensity).

W.r.t. a magnetic field, the Energy density is: $\frac{1}{2\mu_0}B^2$ (μ_0 is permeability & B is Magnetic flux density).

EM wave is made up of electric and magnetic fields, and hence net energy density (u) is:

$u = \frac{1}{2}\varepsilon_0 E^2 +$

But in an EM wave, electric field energy is equal to magnetic field energy, and hence total energy per unit volume or energy density is:

$u = \varepsilon_0 E^2 = B^2/$ $_0$

Also in case of electromagnetic waves, E = cB (c is the speed of light). An improved way to understand energy is to observe how much of energy the wave is carrying from one location to another location. A part of this is wave intensity and it is ratio of the power that crosses an area to the perpendicular area. The energy density "u" and the intensity, "S" are related by a factor "c":$S = cu =$ $+$ $= c\varepsilon_0 E^2 =$

Impact on Health

The fact that electromagnetic fields above some specific values may have biological effects cannot be ruled out. Research has indicated that short-term exposure to values in the environment has no noticeable adverse effects. Human contact to high levels of radiation that could be unsafe is limited by standardized global procedures. The present analysis is to understand whether extended exposure to certain low levels can cause an adverse effect on human health or not [4].Living tissue (sample) of the human body absorbs the energy carried by the electromagnetic radiation [5]. The amount of EM radiation absorbed by the tissue over a period of time is Specific Absorption Rate (SAR). SAR is the foundation for determining the maximal permissible amounts of energy flows from EM fields to which a human being can be exposed. For the whole body given, the mean SAR is given by (σ is the electric conductivity of the sample & "ϱ" is the density (kg/m^3).

Energy assimilated in the fleshy tissue of the active living body can affect a rise in temperature, particularly in case of high frequency fields. Under usual situations, for the absorbed energy if SAR = 4 W/kg, then it prompts an escalation in internal temperature of a person by 1°C. Research has indicated that such an increase has no impact on wellbeing.The limitations on the SAR are the fundamental limitations. According to ICNIRP's (International Commission for Non-Ionizing Radiation Protection) instructions, the conviction is that by constraining the admissible SAR values up to 0.4 W/kg for entire human body, an adequate edge of security at work place can be accomplished. For environmental exposure (general public) the allowed SAR is limited to 0.8 W/kg [6].In India during the year 2008, the Telecommunications Department (DOT) adopted emission guidelines prescribed by ICNIRP to fundamentally curb the levels of electromagnetic radiation w.r.t. antennas (Base Station Emissions) for common public exposure. On the basis of recommendations of the IMC (Inter ministerial Committee), these standards for exposure limit of radiofrequency (emissions from base stations) were strengthened and reduced to one tenth (1/10th) of the limit (according to ICNIRP [8] guidelines). Relevant instructions were given to the mobile network operators on December 30/12/2011. These instructions were further revised on 01/01/2013 and on 06/26/2013(Table: 3).Again, in 2012, EMF exposure limits regulations for mobile towers were revised to one tenth of the existing ICNIRP prescribed limits as a preventative measure, as shown in Table 4 below. At present, the EMF emission limit from the antennas (base station) for exposure to the general public is as indicated in Table 5 below [11]. Also DOT in its letter dated: 27/02/2014 has issued a circular stating that in India, the SAR levels for cell phone handsets to be maintained are 1.6 Watt/Kg averaged over a mass of 1gram of human tissue [11].

Radiation Safety Standards

The entire body exposure limit identified in terms of SAR is 4 W/kg. The wellbeing guidelines are w.r.t. proposals of 2 societies, The United States Federal Communications Commission (US FCC) and The International Commission on Non-Ionizing Radiation Protection (ICNIRP). ICNIRP, a division of W.H.O. (World Health Organization), drafts the principles regarding exposure to electromagnetic fields. ICNIRP data and information covers all Non-ionizing radiation, static and time-fluctuating electric and magnetic fields, RF radiation etc.RF energy absorption by human body alters w.r.t. the frequency of the RF signal. Limiting values for total body exposure are in the 30-300 MHz frequency range (physical body absorbs RF energy more powerfully in this range). The USFCC restricts RF absorption (in SAR terms) to 1.6 W / kg, averaging more than one gram of body tissue for public exposure to cell phone radiation. The permissible limit approved by the ICNIRP is somewhat more, with an average of 2 W/kg over ten grams of body tissue.

Few Preventive measures from Radiation

New technologies have made an ever increasing number of unnatural gadgets producing electromagnetic radiation which are moderately new and concise information on their causative effects on health is not available. It is difficult to demonstrate that any gadget or exposure is completely safe, particularly without such information. To avoid possible dangers, some precautions can be adopted to limit the exposure and impacts of RF radiation like minimizing the duration spent using a mobile phone will minimize exposure to radio frequencies.

- In case a long telephone conversation is inevitable, then it may be helpful to observe a greater physical range (distance) between the physical body and the RF gadget, as the level of exposure decreases with distance.
- Devices with screens protection have been found to lessen a substantial quantity of RF emissions from entering the body.
- Digital cell phones emit less RF radiation than analog phones, reducing potential negative effects.
- Children under the age of 14 should be discouraged from using cell phones and the installation of telecommunications base stations near school and hospital facilities should be discouraged as they along with the patients are more vulnerable to electromagnetic radiation.
- The mobile phone manufacturers must follow necessary safety recommendations while manufacturing them; else the government must intervene when these mobiles produce dangerous levels of RF energy.
- Government's should authorize base station operators to conduct audit on regular basis w.r.t. the prescribed standards for confirming levels of RF exposure to general public exposure.
- All mobile phone manufacturers should provide SAR information on the phone batteries.

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Table 1. Electromagnetic radiation fields and their sources [1]

S.No.	Frequency Field	Range	Sources
01.	Static	0 Hz	Static 0 Hz Natural, Videos (video views), Magnetic Resonance, Industrial electrolysis.
02.	Extremely Low Frequency Fields(ELF)	0-300 Hz	Electric power lines, Electrical wiring, Electric appliances (hair dryers, shavers etc.)
03.	Intermediate frequency fields(IF)	300Hz-10MHz	Anti-theft devices (at shop exits), Computer screens, CFL bulbs.
04.	Radio frequency fields(RF)	10MHz-300GHz	Mobile Phones, microwave ovens, AM/FM Broadcasting, Portable radio systems.

Table 2:The EMF emission limit from the antennas (base station) for exposure to general public is [7]

Frequency Range	E-Field (Strength) (V/m)	H-Field (Strength) (A/m)	Power Density (Watt/m²)
400-2000 MHz	$1.375f^{1/2}$	$0.0037f^{1/2}$	$f/200$
2GHz-300GHz	61	0.16	10

(f- Frequency in MHz)

Table 3: The maximum EMF exposure restriction from antennae (Base station) to general public is [9]

Frequency Range	E-Field (Strength) (V/m)	H-Field (Strength) (A/m)	Power Density (Watt/m²)
400-2000 MHz	$0.434f^{1/2}$	$0.0011f^{1/2}$	$f/2000$
2GHz-300GHz	19.29	0.05	10

(f- Frequency in MHz)

Table 4: Reviewed Electromagnetic Radiation Rules for Cell phone Towers in India [10]

Frequency(MHz)	ICNIRP Radiation Norms	Revised DOT Norms(w.e.f. 01/09/2012)
900	4.5 watts/m²	0.45 watts/m²
1800	9 watts/m²	0.90 watts/m²
2100	10.5 watts/m²	1.05 watts/m²

Table 5

Frequency Range	E-Field (Strength) (V/m)	H-Field (Strength) (A/m)	Power Density (Watt/m²)
400-2000 MHz	$0.434f^{1/2}$	$0.0011f^{1/2}$	$f/2000$
2Ghz-300GHz	19.29	0.05	1

RESEARCH ARTICLE

Synthesis, Characterization and Biological Evaluation of Benzothiazole Derivatives a Potential Inhibitor of COVID-19 Corona Virus Infection by Molecular Docking Studies

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ABSTRACT

In the present work, benzothiazole derivatives have played a vital part in the development of heterocyclic compounds. During the last two decades, the study of the biological activities of benzothiazole derivatives has been the aim of many researchers. The reaction of benzoicacid compounds with ammonium thiocyanate and bromine in glacial acetic acid yield 4-thiocyanatobenzoicacid. The title compounds were synthesized by treating 4-thiocyanatobenzoicacid with o-amino thio phenol and carbon disulphide. Their structures were characterization are done by different analytical techniques, such as elemental analysis, FT-IR, ^{1}H-NMR, ^{13}C-NMR, and ES-Mass. All the synthesized compounds were tested for their anticancer activity against HeLa human cervical cancer cell line with MTT assay. Antimicrobial activity against antibacterial and antifungal activities, anti-inflammatory activity and analgesic activity were studied for the synthesized compounds. COVID-19, a member of corona virus family is spreading its tentacles across the world due to lack of drugs at present. Associated with its infection are cough, fever and respiratory problems causes more than 20% mortality worldwide. It is caused by a positive, single stranded RNA virus from the enveloped corona viruse family. However, the main viral proteinase (Mpro/3CLpro) has recently been regarded as a suitable target for drug design against SARS infection due to its vital role in poly proteins processing necessary for corona virus reproduction. COVID-19, a member of corona virus family is spreading its tentacles across the world due to lack of drugs at present.

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Associated with its infection are cough, fever and respiratory problems causes more than 15% mortality worldwide. It is caused by a positive, single stranded RNA virus from the enveloped corona viruse family. However, the main viral proteinase (Mpro/3CLpro) has recently been regarded as a suitable target for drug design against SARS infection due to its vital role in poly proteins processing necessary for coronavirus reproduction. Docking studies of the synthesized compounds was done with the help of HEX 6.1software using GRIP batch docking method to study their observed activity. Docking study was done and the compounds were found to fit well with the target protein the crystal structure of COVID-19 main protease in complex with an inhibitor N3 (PDB ID: 6LU7).

Keywords: benzothiazole, anti-cancer, anti-microbial, Docking, and anti-inflammatory.

INTRODUCTION

Introduction COVID-19 is easily transmissible and it has already been spread worldwide. Symptoms are flu-like and can include fever, muscle and body aches, coughing, and sore throat. Symptoms may appear 5-6 days after infection. As of March 20h, 2020, over 243,000 cases of COVID-19 have been confirmed worldwide, over 10,000 of which have resulted in death. At present, no specific therapies for COVID-19 are available and research regarding the treatment of COVID-19 are infancy. However, the measures that have been implemented remain limited to preventive and supportive therapies, designed to prevent further complications and organ damage [Morales et al., 2020]. Some preliminary studies have investigated potential combinations that include anti-malarial drug chlorouinone, and anti-HIV vaccines can be used to treat COVID-19 infections. A separate investigation performed by Lu (2020) indicated that among 4 tested drugs (nelfinavir, pitavastatin, perampanel, and praziquantel), nelfinavir was identified as the best potential inhibitor against COVID-19.

The main protease (Mpro)/chymotrypsin like protease (3CLpro) from COVID-19, represents a potential target for the inhibition of CoV replication [Lu, 2020]. It was observed that genome of CoV encodes two proteins ppla and pplb which are involved in spike, membrane, envelop, nucleoprotein, replicase, and polymerase activity of viruses. This function is performed by main protease (Mpro/3CLpro) (Liu and Wang, 2020). The Mpro has 3 structural domains; domain I (residues 8 - 101) and domain II (residues 102 - 184) both have beta barrel motifs representing chymotrypsin catalytic domain and domain III (residues 185 - 200) with a helical structure participates in dimerization of protein and active enzyme production. Given its vital role in poly protein processing and virus maturation, Mpro is considered to be a suitable target for viral inhibitor development as an approach toward SARS. Heterocyclic compounds have great applicability in pharmaceutics because they have specific chemical reactivity. The majority of synthetic heterocyclic compounds have found widespread use, for example as anticancer agents, anti-tubercular, analeptics, analgesic and hypnotics and also in agricultural field as pesticides, insecticides and weed killers. Various synthetic procedures have been developed and considerable diversity in the ring is achieved. Heterocyclic compounds are enormous, their chemistry is complex and synthesizing them requires great skill. Advances in organic chemistry have made possible the synthesis of complexes molecules. Also stereochemistry is an important process in medicinal chemistry since life is inherently chiral and the drug targets within the body are chiral. As such, they can distinguish between the enantiomers of a chiral drug, so the use of racemic drug is inherently wasteful, since only one enantiomer is ideally designed to interact with its target. Moreover, the existences of the "wrong" enantiomer could create problems if it interacted with a different receptor, could become even fatal. The interesting medicinal activities of heterocyclic compounds have stimulated considerable research work in recent years leading to the synthetic utility. Taking in view of the applicability of heterocyclic compounds, we have undertaken the preparation of heterocyclic compound bearing benzothiazole nucleus. The placement of a wide variety of substituents of these nuclei has been designed in order to evaluate the synthesized products for their pharmacological profile against several strains of bacteria and fungi. Benzothiazole is one of the most important heterocyclic compound, weak base, having varied biological activities and still of great scientific interest now a days.

They are widely found in bioorganic and medicinal chemistry with application in drug discovery. Benzothiazole is a privileged bicyclic ring system. Due to its potent and significant biological activities it has great pharmaceutical importance; hence, synthesis of this compound is of considerable interest. The small and simple benzothiazole nucleus if present in compounds involved in research aimed at evaluating new products that possess interesting biological activities. Benzothiazole moieties are part of compounds showing numerous biological activities such as antimicrobial, anticancer, anthelmintic, anti-diabetic activities etc., they have also found application in industry as anti-oxidants and vulcanization accelerators. Various benzothiazoles such as 2-substituted benzothiazole received much attention due to unique structure and its uses as radioactive, amyloidal, imaging agents and anticancer agents. In the 1950s, a number of 2-aminobenzothiazoles were intensively studied, as the 2- amino benzothiazole scaffold is one of privileged structure in medicinal chemistry and reported cytotoxic on cancer cells. It must be emphasized that combination of 2-aminobenzothiazoles with other heterocyclic is a well-known approach to design new drug like molecules, which allows achieving new pharmacological profile, action, toxicity lowering. Benzothiazoles are fused member rings, which contain the heterocyclic bearing thiazole. Sulfur and nitrogen atoms constitute the core structure of thiazole and many pharmacologically and biologically active compounds. Thiazole is structurally related to thiophene and pyridine, but in most of its properties it resembles to the latter. Thiazole was first described by Hantzsch and Waber in 1887. The basic structure of benzothiazole consist of benzene ring fused with 4, 5 position of thiazole. Owing to the vast and wide applications of benzothiazole and its derivatives in industries and industrial operations, in addition to their applications in the field of pharmacology, as antibacterial, antifungal, antihypertensive, anticancer and anti-inflammatory substances, the advancement and development in the field of benzothiazole chemistry have been recognized as a very important area, in the recent time by scientists in different fields including chemistry, pharmacology and medicine. The findings of the present study will provide other researchers with opportunities to identify the right drug to combat COVID-19.

MATERIALS AND METHODS

All the chemicals and solvents used were of AR-grade obtained from Sigma- Aldrich, Sisco Research Laboratories, Qualingens, Hi-media, nice chemicals, Spectrochem and were used without further purification. All melting points were taken in open capillaries and are uncorrected. Elemental analysis was performed on a Perkin-Elmer analyzer. IR spectral [12] were recorded in KBr on Shimadzu spectrometer, ^{1}H-NMR and ^{13}C-NMR in DMSO-d6 on a Bruker AC-400 spectrometer using TMS as an internal standard. The microorganisms were obtained from National Chemical Laboratory, Pune. Thin-layer chromatography (TLC) was performed on pre-coated aluminium plates (silica gel 60F254, Merck). Plates were visualized by UV light and iodine vapor.

General Procedure For The Synthesis of Thiocyanate (TC1-TC3)

The substituted/ unsubstituted benzoic acid (0.5 mol) was dissolved in acetic acid (125 ml) and the solution was added to the solution of ammonium thiocyanate (1.05mol, 80 g) in glacial acetic acid (250 ml). This solution was cooled to 10-20° C. To this well stirred solution, a solution of bromine (0.5 mol, 25.7 ml) in acetic acid (250 ml) was added drop wise for thirty minutes and the temperature was maintained below 20°C. After the addition of bromine, it was kept at room temperature for ten minutes and then it was diluted with an equal amount of water. The solid material was filtered, washed, dried and recrystallized from ethanol.

Compound TC-1 4-Thiocyanatobenzoicacid

Anal. Calcd. For $C_8H_5SN_3O_2$: C, 53.62; H, 2.81; N, 7.82; O 17.86; S 17.82 Found: C, 53.60; H, 2.88; N, 7.88; O, 17.87; S, 17.86; Yield %(76), ES (+) 179 (M+H); M.p.: 173–174 °C; IR KBr (cm $^{-1}$): - ^{v}C≡N: 2220cm-1.

Compound TC-2 2-Chloro-4-Thiocyanatobenzoicacid

Anal. Calcd. For $C_8H_4ClSNO_2$: C, 44.98; H, 1.89; N, 6.56; O 14.98; Cl, 16.59; S 15.01 Found: C, 44.07; H, 1.94; N, 6.60; O, 15.05; Cl, 16.60; S, 15.04; Yield %(62), ES (+) 213 (M+H); M.p.: 199–201 °C; IR KBr (cm $^{-1}$): - ^{v}C≡N: 2231cm-1.

Compound TC-6 3-Nitro-4-Thiocyanatobenzoicacid

Anal. Calcd. For $C_8H_4SN_2O_4$: C, 42.86; H, 1.80; N, 12.50; O 28.55; S 14.30 Found: C, 42.90; H, 1.84; N, 12.56; O, 28.59; S, 14.34; Yield %(75), ES (+) 224 (M+H); M.p.: 172–173 ℃; IR KBr (cm -1): - $^\nu C\equiv N$: 2210cm-1.

General Procedure For The Synthesis Of Benzothiazoles (Compound BT1-BT3)

A mixture of thiocyanate BT1-BT3 (0.01 mol), o-amino thio phenol (0.01 mol) and carbon disulphide (0.1 mol, 8 ml) was heated in an oil bath at 180℃ for 8 hours. The resultant benzothiazole was cooled and recrystallized from ethanol.

Compound 4-(Benzo[D]Thiazol-2-Ylthio) Benzoic Acid

Anal. Calcd. For $C_{14}H_9S_2NO_2$: C, 55.24; H, 2.98; N, 09.16; O 10.50. Found: C, 58.52; H, 03.16; N, 12.32; O, 11.35; S, Yield %(79), ES (+) 287 (M+H); M.p.: 253–254 ℃; ^1H NMR [DMSO-d6, ppm]: δ 7.9 (Ar-H, multiplet), δ 12.40 (Ar-OH, singlet)ppm; ^{13}C-NMR [DMSO-d6, ppm]: δ 169 (OH); IR KBr (cm -1): 1613.19 (C=Nstr), 1530.18 (C=Cstr), 2924.80 (OH str). 722.36 (C-S str) 3080.70 (CH str).

Compound 4-(Benzo[D]Thiazol-2-Ylthio)-2-Chlorobenzoic Acid

Anal. Calcd. For $C_{14}H_8S_2NO_2Cl$: C, 48.13; H, 2.60; N, 08.12; O 12.16. Found: C, 49.82; H, 03.17; N, 10.20; O, 13.23; S, Yield %(80), ES (+) 321 (M+H); M.p.: 289–290 ℃; ^1H NMR [DMSO-d6, ppm]: δ 7.8 (Ar-H, multiplet), δ 12.52 (Ar-OH, singlet)ppm; ^{13}C-NMR [DMSO-d6, ppm]: δ 174 (OH); IR KBr (cm -1): 1601.11 (C=Nstr), 1561.28 (C=Cstr), 2924.99 (OH str). 717.36 (C-S str) 3070.10 (CH str). 781 (C-Cl str).

Compound 4-(Benzo[D]Thiazol-2-Ylthio)-3-Nitrobenzoic Acid

Anal. Calcd. For $C_{14}H_8S_2N_2O_4$: C, 50.1359; H, 2.49; N, 08.43; O 19.16. Found: C, 51.42; H, 02.99; N, 10.16; O, 21.89; S, Yield %(82), ES (+) 332 (M+H); M.p.: 301–302 ℃; ^1H NMR [DMSO-d6, ppm]: δ 7.9 (Ar-H, multiplet), δ 13.04 (Ar-OH, singlet)ppm; ^{13}C-NMR [DMSO-d6, ppm]: δ 165 (OH); IR KBr (cm -1): 1691.23 (C=Nstr), 1592.28 (C=Cstr), 2928.19 (OH str). 684.26 (C-S str) 3090.30 (CH str).1571 (C-NO$_2$ str).

Biological Evaluation

Cytotoxic Activity

Growth of breast cancer cells was quantitated by the ability of living cells to reduce the yellow MTT to purple formazan products. The amount of formazan product formed is directly proportional to the number of living cells. Synthesized compounds were prepared as 4.0 mM top stock solutions, dissolved in DMSO. HeLa human cervical cancer cell line were cultivated at 37°C in an atmosphere of 5% CO2 in Dubecco's modified Eagle's minimal medium (DMEM) supplemented with 3.0 mM L glutamine with 10% fetal bovine serum were routinely sub cultured twice weekly to maintain in continuous logarithmic growth. Cells were trypsinized for the passage into the well plate and plated at 10,000 cells/well in 100 μL of medium in 96-well plates. Cells were allowed to adhere to the surface of well plates. After 24 h, medium was removed and 100 μL of drug solutions (prepared at 6.25, 12.5, 25, 50 and 100 μM concentrations) were added into the wells. 100 μL of fresh medium without cells was added as control. 4 wells were used for each concentration of drug solution, while 4 wells were reserved for cell culture control, which contained the corresponding amount of DMSO. The total drug exposure was 48 h. After 48 h, contents of the well were removed and 20 μL of MTT solution (5 mg in 1 mL of phosphate buffer saline) was added to each well. Incubation at 37°C for 4 h allowed reduction of MTT by mitochondrial dehydrogenase to an insoluble formazan product. Well contents were removed and the formazan product was solubilized by addition of 100 μL DMSO. The purple colour was produced. Absorbance of each well was read on Tenac 200 plate reader at 570 nm. From the absorbance, the % inhibition was calculated as % growth inhibition = Ac – At / Ac X 100.Where *Ac* is the mean absorbance of control and *At* is the mean absorbance of test (Table 1). Nonlinear regression graph was plotted between %cell inhibition and log concentration and IC50 was determined using GraphPad Prism software.

Anti-Microbial Activity

The anti-microbial activity for the sample was carried out by Disc Diffusion Technique [14]. The test microorganisms *Staphylococcus aureus (NCIM 2079), Bacillus subtilis (NCIM 2063), Escherichia coli (NCIM 2065), Klebsiella aerogenes (NCIM 2098), Pseudomonas aeruginosa (NCIM 2036), Candida albicans (NCIM 3102), Aspergillus Niger (NCIM 105)* maintained by periodical subculturing on nutrient agar and sabouraud dextrose agar medium for bacteria and fungi respectively. The test microorganisms were obtained from National Chemical Laboratory NCL), Pune and maintained by periodical sub culturing on nutrient agar and sabouraud dextrose agar medium for bacteria and fungi respectively. The effects produced by the sample were compared with the effect produced by the positive control (Reference standard ciprofloxacin 5 μg/disc for bacteria; Nystatin 100 units/disc for fungi).

Anti Inflammtory Activity

Carrageenan induced hind paw edema

Albino rats of either sex weighing 150-200gms were divided into six groups of six animals each. The dosage of the drugs administrated to the different groups were as follows: Group 1 – Control received normal saline, Group 2 to 4 received test in a dose of 50 mg/kg and Group 5-Indomethacin(10mg/Kg).All the drugs were administrated orally. After one hour of the administration of the drugs, dose 0.1 ml of 1% w/v carrageenan solution in normal saline was injected into the subplantar tissue of the left hind paw of the rat and the right hind paw served as the control. The paw volume of the rats were measured in the digital plethysmograph(Ugo basile, Italy) at the end of 0, 60, 120 and 180 min.The increase in paw edema of the treated group was compared with that of the control and the inhibitory effect of the drugs were studied. The relative potency of the drungs under investigations were calculated based upon the percentage inhibition of the inflammation.

$$\text{Percentage Inhibition} = \frac{\text{Control (increase in paw volume in 3}^{rd}\text{ hour)} - \text{Test(increase in paw volume in 3}^{rd}\text{ hour)}}{\text{Control (increase in pawvolume in 3}^{rd}\text{ hour).}} \text{ X } 100 \text{ ----- } 1$$

Analgesic Activity Method

The analgesic activity of given sample was evaluated by using Hot plate method. The albino mice of either sex were used, the animals were divided into nine groups, 5 animals each. Group 1 control received normal saline (1 ml/kg), group 2 to 4 received the given extract (50 mg/kg orally) group 5 Standard (Pentazocine 10 mg/kg intraperitoneally). Before administering the drug, the basal reaction time was studied by placing the animals in hotplate and the parameter such as paw licking and jumping response were noted. The maximum cut-off time is 15 seconds. After half an hour of administration of the drug as above-mentioned, the reaction time was noted and compared.

Docking Studies (Software Details)

Docking studies of compounds (BT1- BT3) have been performed using HEX 6.1 software which is an interactive molecular graphics program for the drug – VIRAL PROTEIN binding interaction. The structure of the complexes were sketched by CHEMSKETCH (http://www.acdlabs.com). The crystal structure of COVID-19 main protease in complex with an inhibitor N3 (Severe acute respiratory syndrome coronavirus 2, synthetic construct) (PDB ID: 6LU7) was downloaded from the protein data bank (http://www.rcsb.org.pdb). Visualization of the docked poses has been done by using PyMOL software.

RESULTS AND DISCUSSION

The thiocyanates (TC1-TC3) were synthesized in good yield by the reaction of benzoicacid derivatives with ammonium thiocyanate and Br$_2$/CH$_3$COOH under ice-cold condition. Compounds TC1-TC3 on reaction with o-

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amino thio phenol in the presence of carbon disulphide afforded compounds BT1-BT3 Figure.2 Both the ligand are soluble only in DMF and DMSO.

IR Spectral Analysis

The purity of all the synthesized compounds were confirmed by their sharp melting points (uncorrected) and column chromatography. Vibration spectroscopy serves as an important tool to confirm the structure of the compounds. Figure.3 represents the IR spectra of the compounds. The compound shows the characteristic C = N stretching peak at 1613 cm−1, which confirms the formation of the compounds. The compounds (BT1-BT3) also exhibits a series of peaks at 3080 cm−1 (C-H str), 2924.80 cm−1 (OH arom str), 1530.12 cm−1 (C=C str). The infrared spectra of the benzothiazole derivatives BT1-BT3, reveal the presence of bands at 722.23, 717.25 and 684.12 cm-1 respectively, indicating the presence of C-S bond in all these benzothiazole derivatives.. The results provide the required bands characteristics of these benzothiazole compounds. Thus the infrared spectra of these derivatives convincingly lend support to the assigned structures.

^{1}H and ^{13}C NMR Spectroscopy

To further confirm the formation of the compounds (BT1-BT3) the ^{1}H and ^{13}C NMR spectra of the compounds in the absence and presence of were recorded at room temperature (25 °C) in DMSO-d6 with TMS as internal standard (Figure.4 and Figure.5). TMS was used as an internal standard. The ^{1}H NMR spectrum (Figure.4) of the Compound 4-(benzo[d]thiazol-2-ylthio) benzoic acid shows two signals at $\delta(CH_{2-a})$ 7.1, 7.8 and $\delta(OH)$ 12.40 ppm indicating the presence of CH bonds in the derivative. This confirms the formation of the compounds (BT1-BT3) by condensation of carboxylic -OH.

In the ^{13}C NMR spectra of the compounds (BT1-BT3) two peaks appeared at δ 174 ppm which is consigned to -C-OH, respectively, supporting the formation of the compound. In the ^{13}C NMR of the compounds (BT1-BT3) these peaks were shifted to lower and higher value [OH] δ 169ppm .The peaks due to other protons remained unaltered which proves their non-involvement in coordination. Both the ^{1}H and ^{13}C NMR spectral studies confirms the formation of the compounds

ESI Mass Spectroscopy

The formation of the compound BT1-BT3 and their proposed structure was further confirmed by the ESI mass spectra. The spectra of the compound BT 3(4-(benzo[d]thiazol-2-ylthio)-3-nitrobenzoic acid) (Figure.6) show its molecular ion peak at m/z 332 which is in good agreement with the formula weight. The spectrum also exhibited a series of peaks at m/z 332,306, 283, 248,187,156, 122, 78 which can be attributed to its further fragments. It is worth mentioning that the results of mass spectrum are in agreement with micro analytical data and suggested formula. The coming part of the discussion deals with the mass spectrum of 4-(benzo[d]thiazol-2-ylthio)-3-nitrobenzoic acid (BT3). On electron bombardment, the benzothiazole compound BT3 is found to lose an electron to form a radical cation, which ejects a molecule of acetylene, resulting in the formation of the base peak, m/z 187 (100 %). The following scheme (Scheme 2) represents the formation of various fragments:

Cytotoxic Activity of BT1-BT3 Compounds

The cytotoxicity assays of BT1-BT3 compounds against human cervical cancer cell line HeLa were conducted at five different concentrations (100, 50, 25, 12.5 and 6.5 μg/mL) for 24 hours, and the results are shown in Figure.8,9&10. The results showed that both the compounds inhibited the growth of the HeLa cells and their effect found to be concentration and time dependent. The concentrations required to inhibit the growth of 50 % of the cells, i.e. IC50 value was calculated for the sample against HeLa cell lines. The IC50 values for BT2 compound (14.35μg/ml) is higher than for BT3 and BT1 compounds (17.25 μg/ml and 22.71 μg/ml). This can be attributed to structural aspect of the compounds. The higher planarity of BT2 compound enabled to penetrate through the cell walls HeLa cells more than BT3 and BT1 compounds. It is also noteworthy that both the compounds were more effective than cis-platin at reducing the proliferation of the HeLa cells.

Antimicrobial Activity

Much attention is being paid to the development of safer antioxidants because of their vital role in food, cosmetics and pharmaceutical products [43] and in the prevention of diseases caused by oxidative stress. The synthesized compounds (BT1-BT3) and the licensed antimicrobials have been tested for their in vitro antimicrobial activity against the bacteria (Staphylococcus aureus (NCIM 2079), Bacillus subtilis (NCIM 2063), Escherichia coli (NCIM 2065), Klebsiella aerogenes (NCIM 2098), Pseudomonas aeruginosa (NCIM 2036), Candida albicans (NCIM 3102), Aspergillus Niger (NCIM 105). The minimum inhibitory concentration (MIC) was determined by broth micro dilution by examining serial dilution of concentrations along with the standards at the same concentrations. The results of the tested compounds are summarized in Tables 2. Effect of the solvent (DMSO) on microbial growth was tested and found that the solvent has negligible effect on bacterial growth. From the results, it is clear that the compounds (BT1-BT3) have higher inhibitory effect than the free BT3 and the standard drugs. This is because of change in structure due to coordination and chelating effect which in turn make the compounds to act as more potent antibacterial agents, thus killing or by inhibiting multiplication of the microbe by blocking their active sites [44]. Such increased activity of the metal chelates can be explained by the reduced polarity of the compounds due to the overlap of the compounds orbital and partial sharing of the positive charge of the metal ion with electron releasing groups. It is evident that reducing the total electron density on free ligands makes the diffusion proceed faster through the bacterial cells [45]. However, BT3 compound exhibited a higher antimicrobial activity than BT2 and BT1 the significant antimicrobial activities of both the compounds (BT1-BT3) may be related with their DNA binding affinities.

Analgesic Activity Study

Hot Plate Method

The analgesic activity of the synthesized compounds was determined at 50 mg kg^{-1} body weight of the animal in the hot plate method. The results suggest that these compounds possess a fairly good analgesic activity. A critical examination of the data in Table 3 clearly indicates that the analgesic activity of the test derivatives is less than that of the standard drug, pentazocine. Moreover, compound comprising BT1 shows the best activity among the synthesized compounds. The Table 3 presents the results of the study of the effect of the drug on the analgesic activity by Hot Plate Method. The relevant data are represented in the figure (Figure.13)

Anti-Inflammatory Activity Study

The anti-inflammatory activity of the synthesized derivatives was determined at the dose level of 50 mg kg^{-1}body weight of the animal, albino rat. The results were compared with the anti-inflammatory activity of the standard drug, indomethacine. The percentage of inhibition of paw edema was calculated. The results are furnished in the Table 4. The compound BT 2 again shows the maximum activity. This order agrees closely with the order observed in the other studies too. The Table 4 presents the results of the study of the effect of the drug on the anti-inflammatory activity by Carrageenan induced hind paw oedema method. The relevant data are presented in the figure (Figure.14).

Docking Studies

The interaction residues and energy values of the synthesized compounds with the target.

Molecular docking with VIRAL PROTEIN

Molecular docking technique allows us to understand the interaction between a drug and protein at the molecular level. In the present study, molecular docking of the compounds   (BT1- BT3) with protein the crystal structure of COVID-19 main protease in complex with an inhibitor N3 (Severe acute respiratory syndrome coronavirus 2, synthetic construct)   (PDB ID: 6LU7) was performed in order to rationalize the mode of protein and most favourable binding conformations of the molecules. Figure15 shows the minimum energy docked pose of the compounds (BT1–BT3) from the results it is clear that, both the compounds (BT1-BT3) interact with VIRAL PROTEIN via intercalation mode of binding. This could be explained by the fact that, stacking interaction of compounds (BT1-BT3) with oxygen atom of the phosphate backbone leads to the formation of stable complex as reported in literature [27]. The resulting

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relative binding energy of compounds (BT1–BT3) with VIRAL PROTEIN were found to be -233.78,-243.18 and -251.12 KJ mol−1, respectively, The results of the docking view revealed the fact that the complexes bind with protein via intercalation and that the complexes stabilize the protein by van der Waal's and hydrophobic interaction. It is also to be noted that the compounds exhibit more binding affinity than compounds (BT1-BT3). The binding energy of the compounds follows the order BT2 > BT3> BT1 which is in good agreement with the binding constants obtained from absorption and emission spectral study

CONCLUSIONS

The present investigation is focused on the synthesis, characterization and biological activities of a series of benzothiazole compounds from 4-thiocyanatobenzoicacid.Three thiocyanates (TC1-TC3) were prepared from substituted/unsubstituted benzoicacid. Further, Three compounds, BT1-BT3 were synthesized, following the scheme (Figure 2). The infrared spectra of all these compounds provide the expected frequencies. The ^{1}H NMR and ^{13}C NMR spectra of all the seven compounds provide the expected signals. The mass spectra of all the compounds were recorded. Probable schemes were provided for fragmentation patterns. All the compounds were tested for their anticancer activity against HeLa human cervical cancer cell lines with MTT assay. Most of the compounds showed moderate to good anti- cervical cancer activity. LD_{50} values were determined and these are reported. The anti-inflammatory activity study showed that compound B1 has significant effect over carrageenan induced hind paw edema. On percentage protection basis, compound B2 showed 42.25%, while Indomethacin showed 63.80% when compared to control. Compound B1 proved to possess potential analgesic activity. A study of the antimicrobial activity was carried out and the results are given. Due to non-approved drugs at present Currently, COVID-19 has emerged in the human population, in China, and is a potential threat to global health, worldwide. Currently, the main target for COVID-19 treatment primarily act on the main protease (Mpro). The aim of this study was to examine Benzothiazole that may be used to inhibit the COVID-19 infection pathway. Benzothiazole has high binding affinity and lowest binding energies. Therefore, we suggested that Benzothiazole may represent potential treatment options, and found in medicinal chemistry that may act as potential inhibitors of COVID-19 Mpro. However, further studies should be conducted for the validation of these compounds using in vitro and in vivo models to pave a way for these compounds in drug discovery.9

ACKNOWLEDGEMENT

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Table 1 MTT assay measurement of various percentages of cell deaths for different concentrations of benzothiazole derivatives

Compound	% Growth inhibition				
	6.25 µM	12.5 µM	25 µM	50 µM	100 µM
BT 1	0.48	3.71	8.53	22.71	22.25
BT 2	0.45	0.04	2.45	14.35	36.70
BT 3	0.43	1.25	4.25	17.25	37.25

Tables 2 Antimicrobial activities of the synthesized compounds

Name of the Microorganisms	DMSO Extract 100 µl added and Zone of inhibition (mm/ml)			
	BT1	BT2	BT3	Control
Staphylococcus aureus	12	15	20	28
Bacillus subtilis	18	15	27	30
E.coli	18	28	26	25
Pseudomonas aeruginosa	16	20	25	35
Candida albicans	16	14	16	30
A.niger	17	15	25	25

Table 3 Analgesic Activity of the test compounds in Albino mice by Hot plate method

S.No	Groups	Drug	Dose (mg/kg)	Reaction time (in sec)			
				Before Administration of drug	After Administration of drug		
					30(mins)	60(mins)	120(mins)
1.	Control	Saline	1ml/kg	4.41±0.16	4.42±0.10	4.28±0.30	4.43±0.27

2.	Test-1	BT1	50mg/kg	4.16±0.24	8.27±0.16	9.38±0.18	9.52±0.28
3.	Test-2	BT2	50mg/kg	4.22±0.32	7.34±0.18	8.48±0.24	9.31±0.32
4.	Test-3	BT3	50mg/kg	4.46±0.16	8.14±0.18	9.12±0.18	9.36±0.24
5.	Standard	pentazocine	10mg/kg	5.42±0.26	14.6±0.37	16.6±0.18	15.66±0.28

Table 4 Anti-inflammatory activity of test compounds against Carrageenan induced hind paw oedema method in Albino Rats.

Treatment	Dose mg/kg p.o.	Paw volume Increase after 3 hr (ml)	Percentage Inhibition
Control	5 ml/kg	111.61±10.56	-
BT 1	50mg/kg	62.17±6.17	32.89
BT 2	50mg/kg	61.46±4.45	42.25
BT 3	50mg/kg	64.28±4.92	33.44
Indomethacin	10mg/kg	0.4±3.62	63.80

*$P < 0.001$ values are expressed as ± SEM   Number of animals used are 6 in each group

Table 5: Energy values of the synthesized compound

S.No	Compound	Energy
1	BT 1	-233.78
2	BT 2	-243.18
3	BT 3	-251.12

Mpro Protein COVID-19

Benzothiazole

Molecular docking
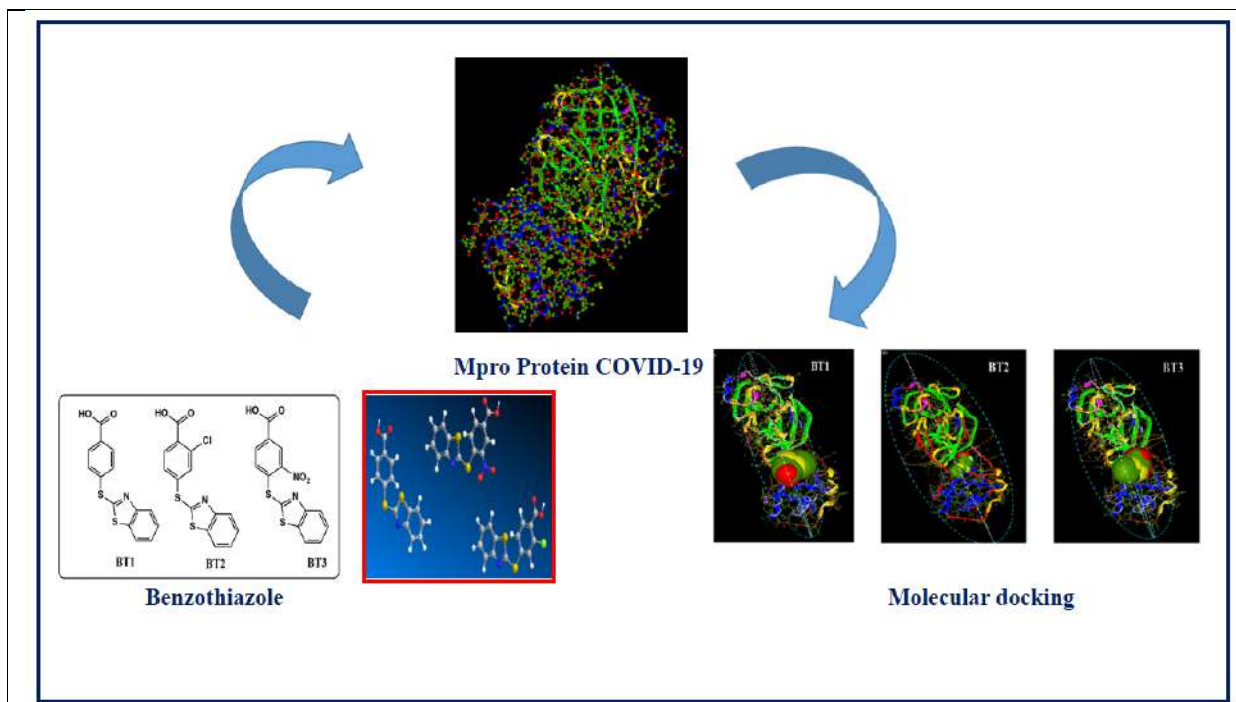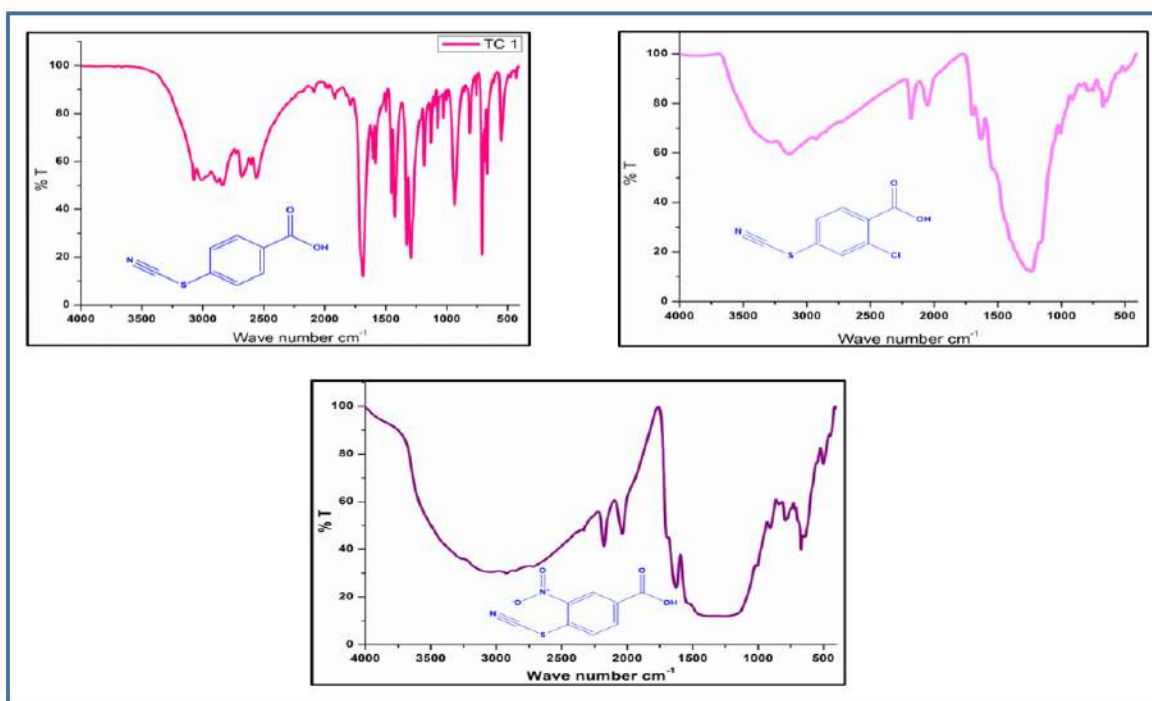
Figure 1. Infrared Spectrum of substituted 4-thiocyanato compounds
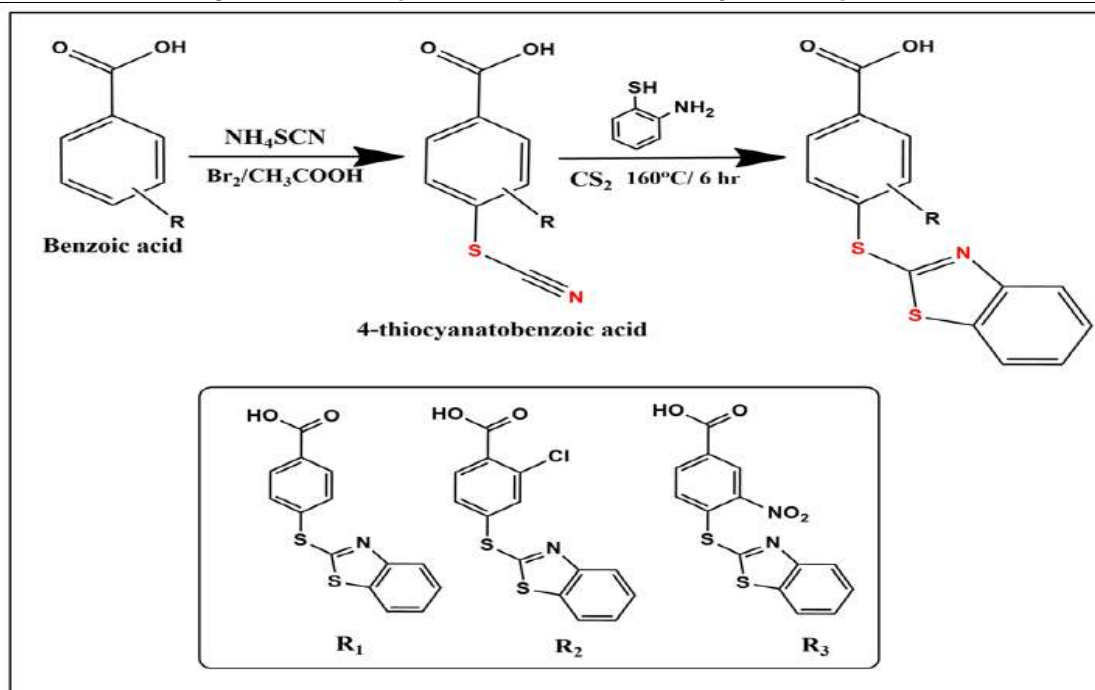
Figure.2 Schematic diagram of synthesis of benzothiazoles (BT1-BT3) compounds

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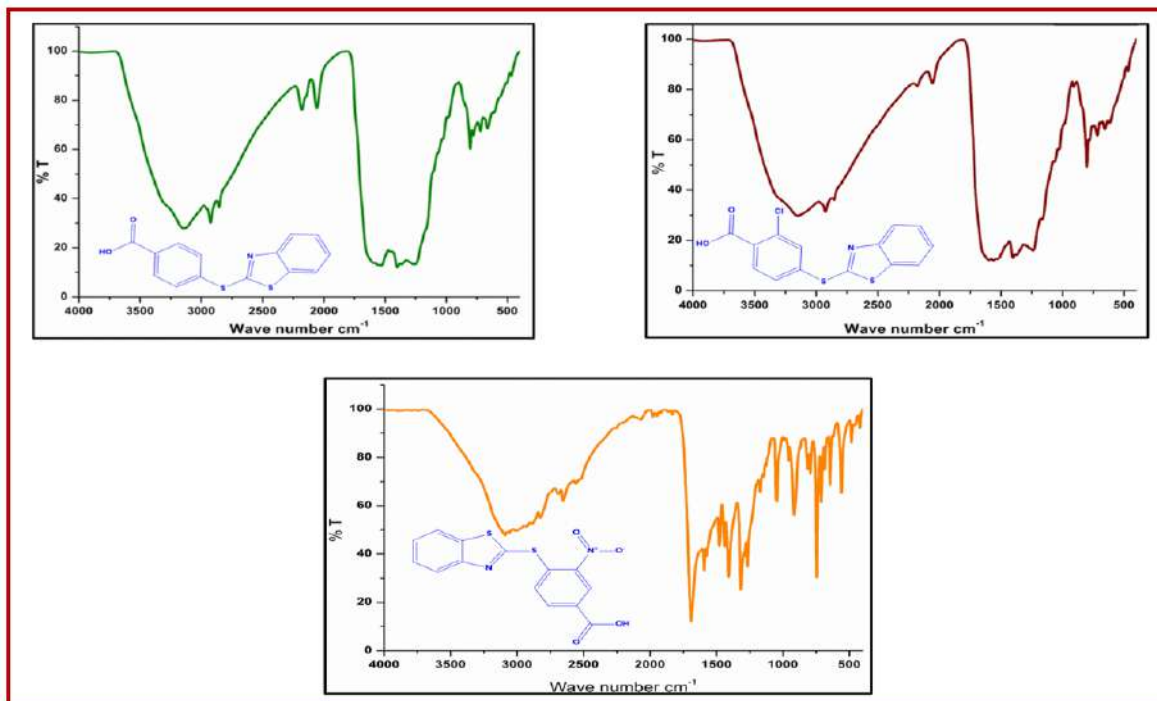
Figure 3. Infrared Spectrum of substituted benzothiazoles (BT1-BT3) derivatives
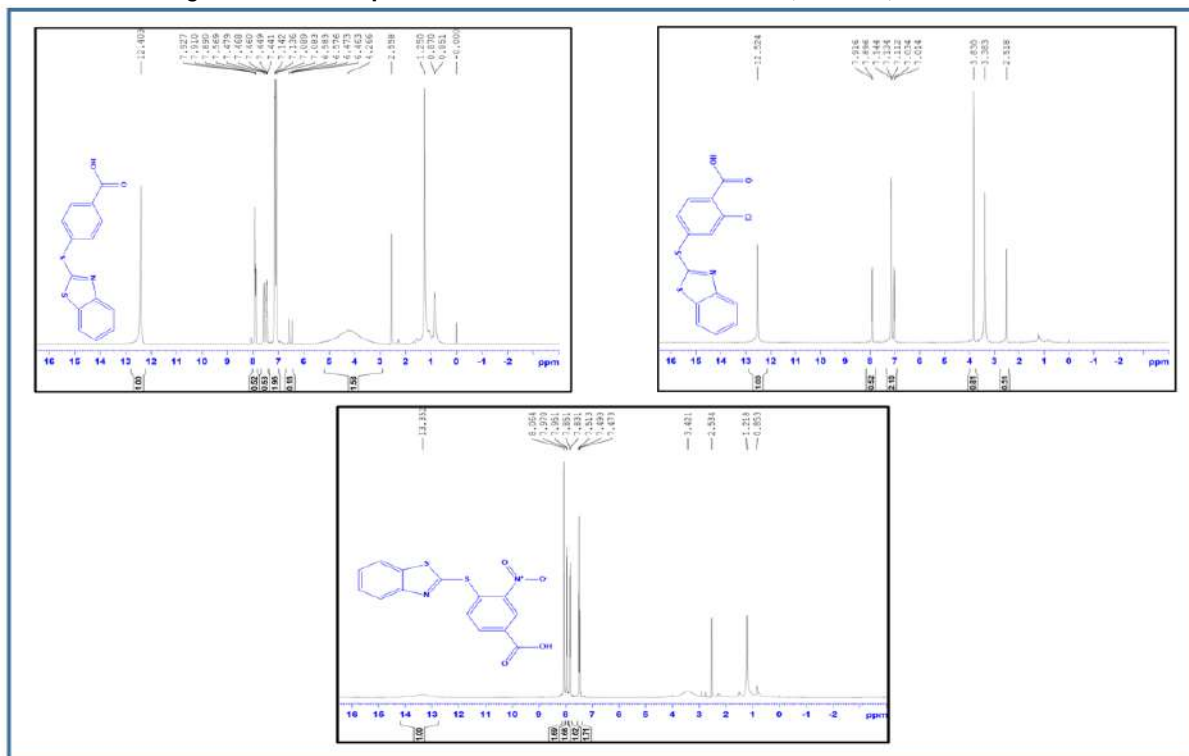
Figure 4. ¹H NMR spectrum of substituted benzothiazoles (BT1-BT3) derivatives

Figure 5. ¹³C NMR spectrum of substituted benzothiazoles (BT1-BT3) derivatives
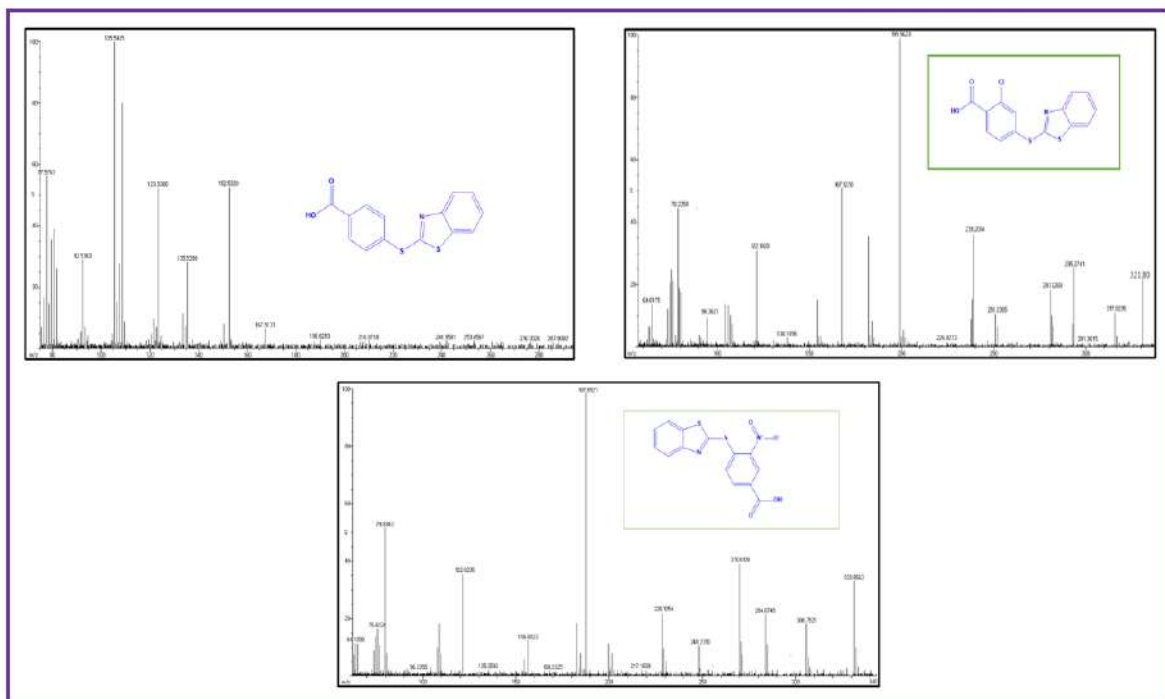
Figure 6. Mass Spectrum of substituted benzothiazoles (BT1-BT3) derivatives

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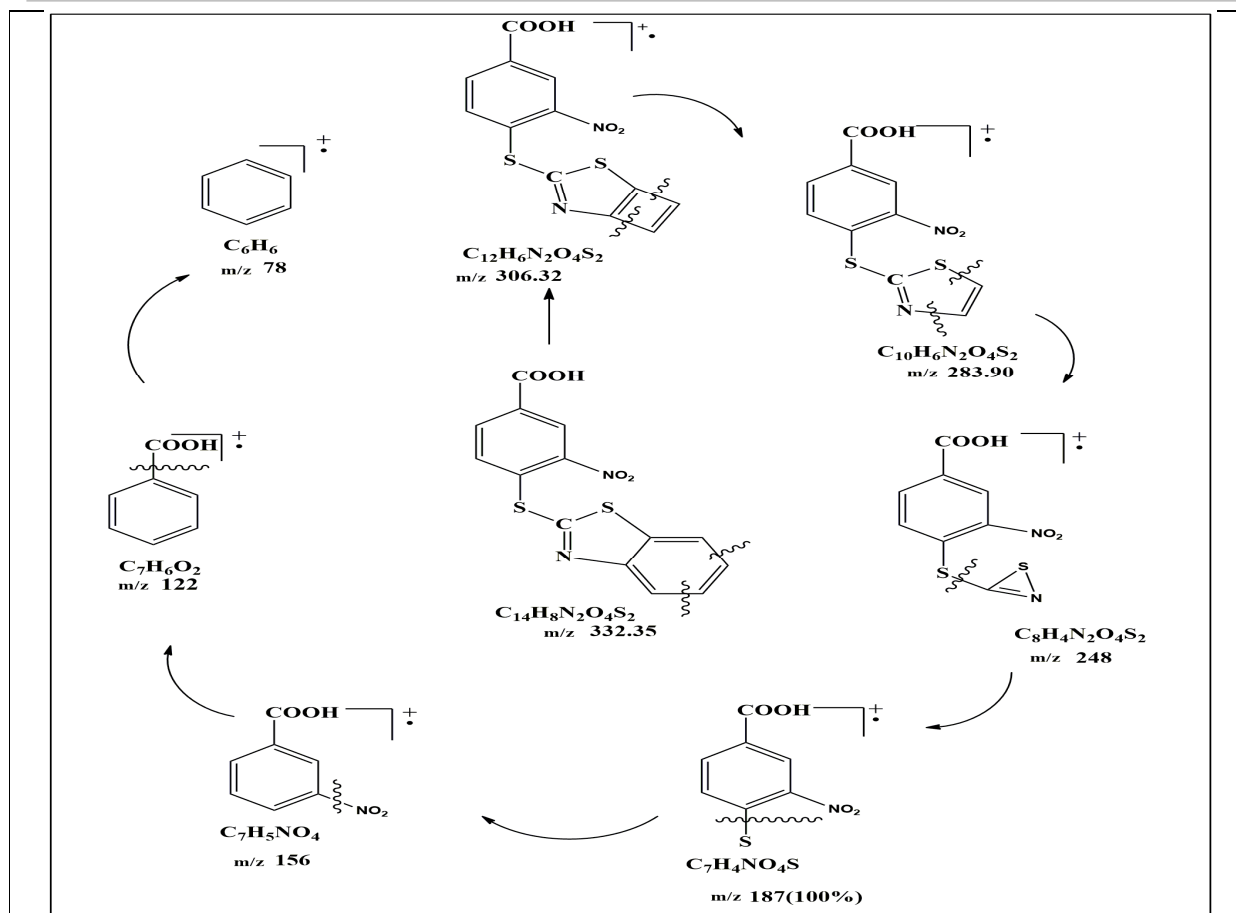
Figure.7 Schematic of mass fragmentation patterns benzothiazole BT 3(4-(benzo[d]thiazol-2-ylthio)-3-nitrobenzoic acid) compounds
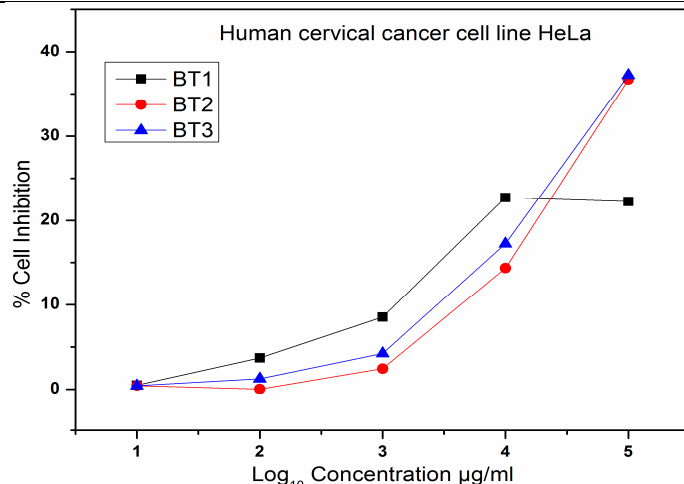
Figure.8 MTT assay measurement on different percentages of cell viability against varied concentrations of BT1-BT3 Compounds

Figure.9 BT1-BT3 Effect on HeLa (human cervical cancer cell lines) morphology upon treatment with various concentrations of benzothiazole derivatives.
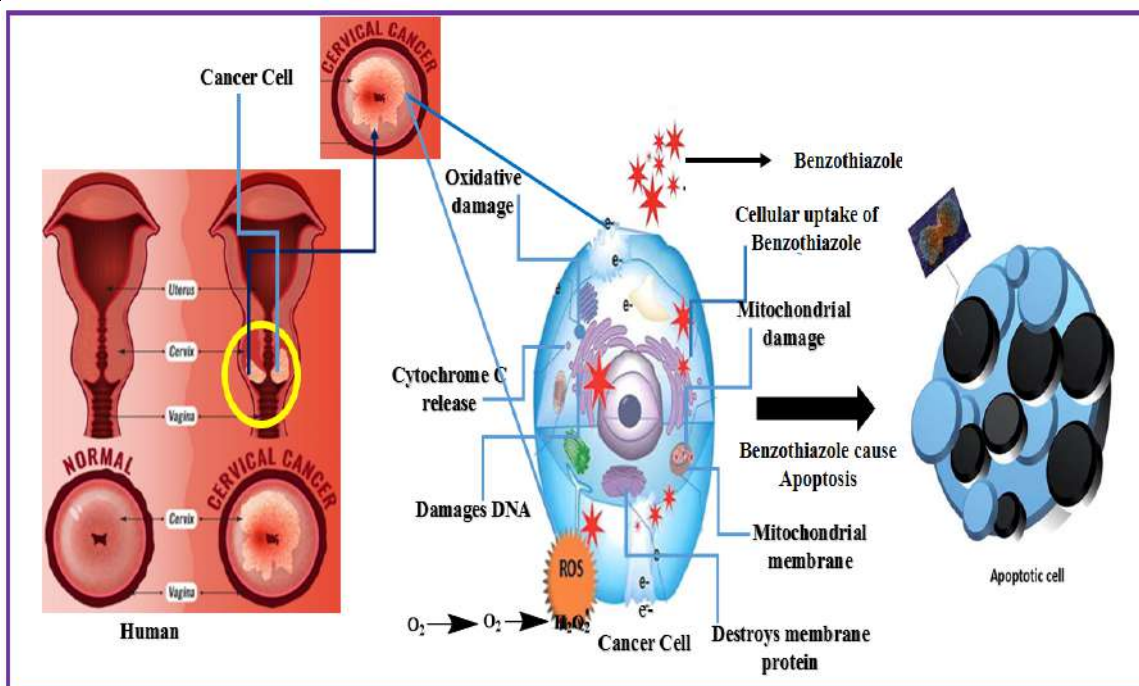
Figure.10 Schematic of cytotoxicity effect of benzothiazole derivatives against HeLa human cervical cancer cell lines

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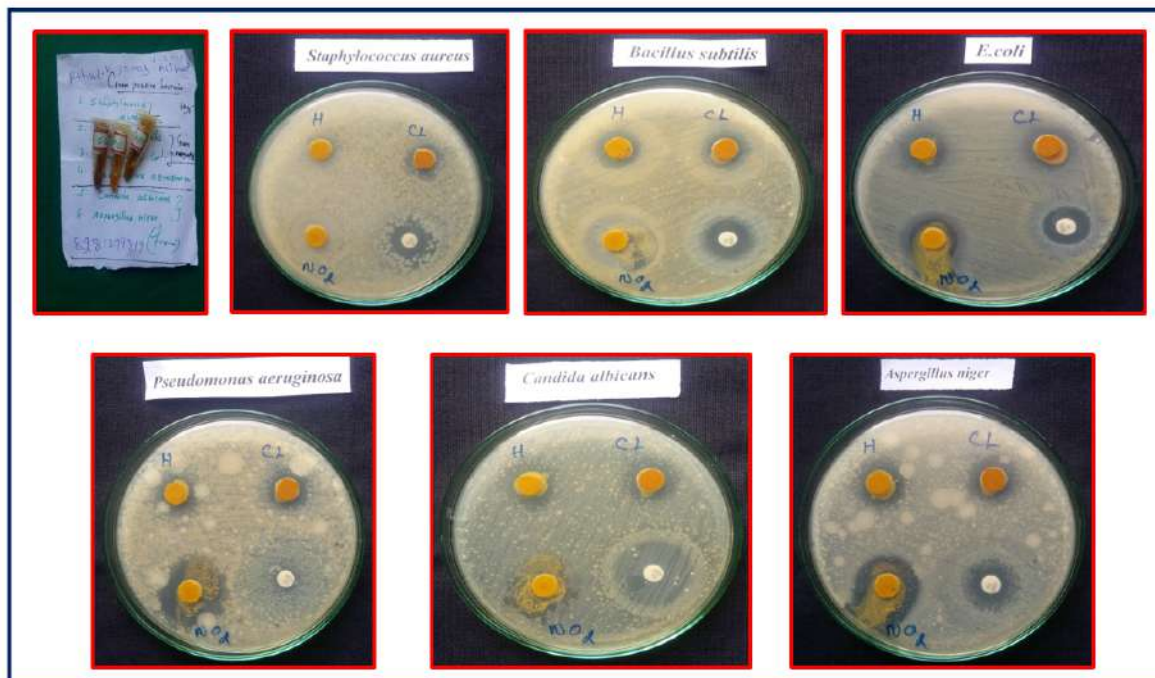
Figure.11 Zone of inhibition given by (BT1-BT3) against bacteria and fungi
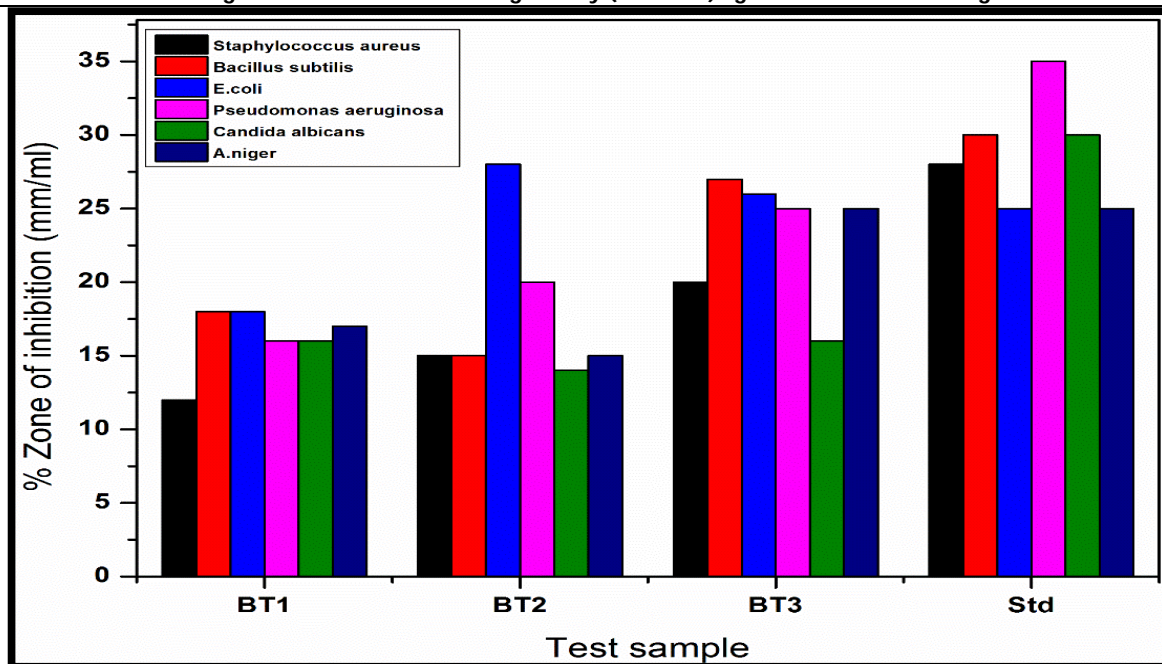
Figure.12 Antimicrobial activity of the synthesized benzothiazoles (BT1-BT3) compounds

Figure.13.Analgesic activity of the synthesized (BT1-BT3) compounds
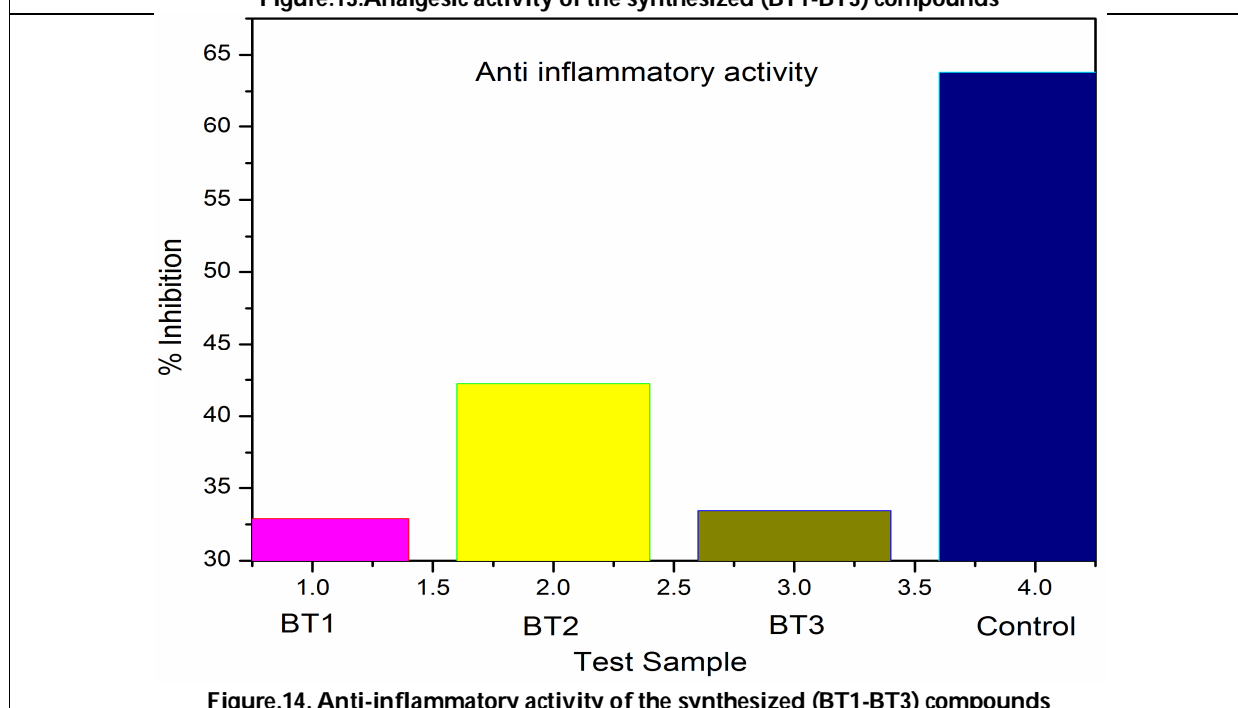
Figure.14. Anti-inflammatory activity of the synthesized (BT1-BT3) compounds

Figure .15 Molecular docked model of compounds (BT 1-BT 3)

RESEARCH ARTICLE

An Empirical Study on Ontology Learning Tools and Applications

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ABSTRACT

Text is represented over Internet in three formats, viz., structured, semi-structured and unstructured. Almost eighty percent of electronic data is in semi or un-structured format that is dynamic in its content and nature. Operations over such huge textual information become tedious and hence information retrieval (IR) becomes a mammoth task. Ever-growing unstructured type of data on internet necessarily calls for a machine-driven approach to deduce and decipher its content in an automated way. Various methods of representing data in a machine-readable format are available. Ontology learning (OL) is one such field that deals with knowledge representation techniques. Semantic web (SW) is powered by efficient techniques using OL methods and tools which support three different types of data for semi-automatic ontology engineering process. Numerous tools for OL from text are available. These tools use distinct methods and approaches. This survey presents an exhaustive survey of thirty OL tools that have come into existence. It reveals that not many tools have emerged in the recent past and that most tools are implemented using techniques of artificial intelligence (AI), knowledge acquisition (KA), machine learning (ML), IR, natural language processing (NLP) etc. Very few tools have blend of more than one technique. Tools and structures for KA, NLP and knowledge management are autonomous from each other, even though a number of components are shared between them. There's more scope for developing tools to convert natural text to semi or partially structured text. More research in the field of pattern-based approaches to ontology construction is needed. The complex nature of ontology development (OD) and its significance for web-based knowledge systems drives this field of research to be relevant. Yet another area of research is evaluation of ontologies which brings about efficient and effective tools. OL tools help reduce development time and cost by extracting knowledge from the texts.

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INTRODUCTION

The Internet with its services has become a vast reservoir of structured, semi-structured and unstructured mine of information. The nature of data available over the web is ever-changing and ever-growing. Hence, web's voluminous data and its volatile nature of content makes retrieval of information a tedious task. Therefore, the ever-increasing volume of unstructured type of data accumulating over the web calls for automated approach to segment, derive meaning, and interpret its contents in a meaningful way, mostly in an automated way. Unfortunately, a lot of stored data is in unsemantic-driven format and hence IR is not machine-processable. The SW, an expansion of World Wide Web (WWW), is aimed to accumulate mesh of data in vocabularies, semantically associated to process by machines both in logical and lexical forms. It is vital to enrich human or non-human interaction with the representation of data in a machine-readable format [1]. Ontology Engineering (OE) deals with the technique of representing data in a machine-readable format [2-8].The present survey is a study on various OL tools that help automate or semi-automate OL from text through various methods, systems and applications. The paper is chapterized as follows: Chapter 2 is a short note on preliminaries on ontology, OL, OL systems and types of ontology tools. Chapter 3 deals with related works while Chapter 4 gives an overview of OL tools. Chapter 5 deals with research issues. Finally, chapter 6 concludes the survey.

Preliminaries
OL from Text
OL from text implies a collection of procedures and techniques for building or enrichment of ontology [9], using diverse types of textual sources. Maedche and Staab categorized OL approaches based on entries such as: "texts, dictionaries, knowledge bases, semi-structured schemas and relational databases" [10]. Each of these methods has explicit technique and related tools. The task of building ontologies from texts basically encompasses extraction of terms, discovery of synonyms, generation of taxonomies, relationships and extraction of rules. The steps are in a layered form, each requiring complex learning technique over the lower one as shown in figure 1 [11]. Extraction of terms is a precondition for other steps in OL from text [12].

Types of Ontology Tools
In the past two decades numerous OL methods and systems have come into existence. Several are autonomous OL systems and some others are tools. Ontology tools (OT) are of various types, which are created to build, edit, annotate, and merge ontologies. Based on objectives of each tool, Gomez, P., categorized OT as: [13, 14]
- Ontology Development Tools
- Ontology Merge and Alignment Tools
- Ontology Evaluation Tools
- Ontology-based Annotation Tools
- Ontology Querying Tools and Inference Engines and
- Ontology Learning Tools.

Related Works
The survey titled "OntoWeb Consortium" [9], shows eighteen tools for OL from text. The tools are presented with their main goals, techniques used, method followed with types of sources, user intervention if any, software architecture, possible interoperability with other tools, and interface facilities. The research article by Shamsfard M., and Barforoush A, [8] is focussed on selecting seven important systems to explain and compare unambiguously among fifty studied works. The criteria for selecting systems were well-built autonomous and assisting tools, possessing distinguishing features and well-documented systems. The authors described the

unearthed feature set to categorize and equate OL systems and compare the selected tools based on the introduced framework.

To construct ontologies and help the ontologist in the acquisition process Gomez-Perez A. and Manzano-Macho D [15] summarized fourteen most relevant tools. They focused on identifying and grouping the tools based on their aim and ontology elements learnt with each tool as three categories: viz., relationship learning tools, concept learning tools, and taxonomies construction tools. Barforush, A., and Rahnama A., [16] arranged OL into four types (terms, taxonomy, ontology, and axioms) and specified their framework. They presented OL applications/tools that were used for creating ontologies from unstructured and semi-structured texts. The authors reviewed various OL applications from the year 2004 to 2012. They are: Bole, OLE, Text2Onto, Englmeier et al's tool, Isolde, Sole, Dynamo, LexO, OntoLancs, Wongetal's tool, OntoCase, ORE, LexOnt, OntoCmaps, AEON, RelExt, ISOLDE, OntoGen, and GALEON.

Wong et al., [17] presented in detail seven well-known OL systems of the foregone decade. The authors examined and evaluated the techniques used by seven OL systems. These systems have wide acceptance and comprehensiveness with regard to many OL tasks and outcomes. Zahra, Faruk Mustafa, et al. [18] intended to present some semi-automatic OL tools from texts, which use diverse techniques and methods. They focused on central features and usability from the view point of a commoner. The authors fixed on only three available OL tools for installation and testing a corpus of fifteen scientific papers in English language on learning ontologies. They also focused on two OL tools for Portuguese language ONTOLP and PORONTO. The authors tested on KEA, Text-To-Onto and PORONTO tools. They concluded that use of tools reduced development time and cost by extracting knowledge from the texts. However, while developing tools with techniques it would always be a human intervention that makes a final decision to include a concept or establish relationship among concepts in the ontology.

Al-Arfaj, Abeer, et al. [19] presented a short summary on linguistic, statistical and hybrid approaches in OL while referencing to main dimensions that classified these methods. OL systems which unearth ontological structures in the given textual sources were summarized. Further, a comparison and a discussion on major distinguishing factors among OL systems (DODDLE II, Text-To-Onto, Text2Onto, OntoLearn, HASTI, OntoGen, TextOntoEx, OntoGain, CRCTOL, OntoCmaps, and LexOnt) was carried out. Their study revealed that many current OL systems depended on light NLP techniques and statistical approaches. The tools employed light NLP techniques and the goal was to extract concepts and taxonomic relations.A distinctive approach in representing knowledge and reasoning for semantic exploration of different kinds of documents is formed by Mishra S, and Sarika J, [4] while presenting concepts and knowledge needed for the intended approach. They surveyed research articles on several approaches for OL in a domain context, which qualified reasoning with indefinite, partial and conflicting information. The authors intended to enable ontological knowledge representation (KR) using texts in real-life situation across numerous knowledge extraction and learning methodologies. They opined three things to do by this exploratory study; 1) retrieve pertinent terms from unstructured or semi-structured data, 2) form concepts and 3) develop the concept hierarchy. These provide the basis to develop ontology for KR framework. [4] They aimed at providing a framework for OL KR in a unique and portable manner.

Fatima N. AL-Aswadi and Chan Huah Yong [20] reviewed various research works in OL systems from text. They elicited defects and noted that many OL methods did not encompass the entire OL process and several methods used shallow architecture for NLP and/or ML. They pointed shallow architectures encountered difficulties when faced with complex real-time applications like human speech, natural language (NL) and natural image. They observed relations discovery results in these methods was still less than satisfactory and advocated deep learning (DL) for relations discovery to enhance results. The authors strongly felt a need to standardize framework for evaluating OL methods. The authors proposed two suggestions to enhance OL process, viz., to use DL methods for i) semantic syntactic parsing and ii) constructing suitable training model by means of pre-processed datasets as input. Somodevilla, M. J., et al., [21] considerably reviewed research works on OL types and tasks. They dealt

with rudimentary types of OL, schema extraction, construction and population, along with measures and evaluation methods. The authors presented seven OL tools whose general objective was to gain semantic knowledge from textual sources for subsequent role in ontology development. The summary of the tools covered motivation, application domain, techniques employed and corresponding tasks to be achieved.

An Overview of Ontology Learning Tools

Tools for OL from Text

In the past two decades automatic and semi-automatic OL tools have come into market. OL tools are designed to enhance learning process in an effective manner. Park, Cho [22] differentiated OL tools into three categories, viz., i) editing ii) merging and iii) extraction. OL extraction tools are further categorized into three types [15] viz., learning relations, new concepts, and supporting in constructing a taxonomy. Learning relations tools help in discovering new relations from the selected input, either taxonomic or non-taxonomic. Tools that aid in acquiring new concepts support in searching and finding new concepts. Tools which assist in building taxonomy help the ontologist to build new ontology and/or enrich an existing one. The OL process that is captured from the tools taken for study is as follows: Domain specific terms are extracted to derive concepts. The OL systems determine synonyms using clustering and applications of statistical similarity measures. Concepts are discovered by grouping the terms that are unique. The arrangement in a taxonomy structure of the extracted concepts is done in the concept hierarchy of OL system with the help of hierarchical clustering. The extraction of anonymous associations or verbs which indicate relationship between the entities is done in during the learning of non-taxonomic relation. The axioms, for the extracted concepts, are created by analysing the syntactic structure of natural language and transformational rules are applied on the subsequent dependency tree. Then the ontology is augmented with instances of concepts and properties grounded on the matching of lexical and syntactic patterns. Instances and properties are added based on tuples collected. The goal of the OL system is to extend the taxonomic structures of a prevailing ontology with further concepts with the application of semantic similarity measures. The OL system attempts to extract complex relations by applying SVM method or SRL method. The observation made from the survey reveals that these steps do not apply in every OL system.

Classification of Tools

The most prominent, available and relevant tools to unearth ontological structures are classified under i) automatic, ii) semi-automatic and iii) non-automatic tools.

Automatic OL Tools

CORPORUM-Ontobuilder: CORPORUM-Ontobuilder is a product of CognIT. It aims to obtain ontologies (primarily taxonomies) from NL texts. The analysis and IE functionalities are driven by various linguistic techniques. OntoWrappper [23] tool extracts information from structured (on-line resources) database while OntoExtract [24] tools acquires arrangements from NL texts. It also provides original concept taxonomies, refines current concept taxonomies, looks for associations among significant terms and concept occurrences within files. Concept taxonomies are built in RDF(S).

Differential Ontology Editor (DOE): DOE [25, 26] ontology editor follows three steps to build ontologies. Firstly, user improves concept and relation classifications in qualifying unambiguously their location in the hierarchy. Secondly, two taxonomies get introduced so the user may add restrictions to domains of relations. And lastly, using XSLT style sheet, the ontology gets converted into a KR language through conceptual graphs or DAML+OIL, RDF(S) and OIL [9]. DOE is more a complement to other editors than intended as OD environment.

Keyphrases Extraction Algorithm (KEA): KEA [27] extracts key phrases from text documents automatically. Lexical processing helps in identifying a group of all candidate phrases. Characteristics are computed for each candidate. The ML generates a classifier to decide which candidates to be designated as key phrases. The ML scheme with known key phrases in training documents, first builds a prediction model, then replicates it to find

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key phrases in new textual sources. KEA's extraction algorithm follows two phases: viz., training and extraction. The efficiency of the method is assessed on how many assigned significant phrases are rightly identified.

SVETLAN': SVETLAN' [8] is developed to categorize nouns in context. Taking into consideration the context in which the words are used, mixing of meanings is avoided. SVETLAN' functions as domain independent tool. Its learning technique is founded on distributional methodology: sentences where nouns with identical syntactic role juxtaposed with identical verbs are combined in the identical class. It follows three-step learning process: 1) syntactic analysis, 2) aggregation and 3) filtering. The syntactic analysis using tools to retrieve sentences from the original texts. The verbs in the retrieved sentences are then found [15]. The assumption is that verbs classify nouns. The result is a group of triplets containing, a syntactic association together with a verb and a noun. Nouns having meanings alike are grouped in aggregation. The weight of the nouns within their classes are considered for filtering. Irrelevant nouns are removed. Since the whole process is completely independent, it doesn't require validation [9].

Welkin: Welkin [28] is designed for automatic generation of e-learning materials from free textual sources. The aim is to generate an ontological representation for terms of interest, and internally represent various sections in the e-learning web sites. Module analyses texts to identify relevant terms, and classify them in lexical ontologies using contexts. Each concept in base ontology is supported with words in context having syntactic relationships among them. Distance metric is used to classify new terms inside ontology. This method automates classification of oft-repeated occurrence of concepts from ancient sources to create e-learning web sites.

Concept-Relation-Concept Tuple-based Ontology Learning (CRCTOL): CRCTOL [29], handles linguistics and statistics-based procedures to accomplish its OL tasks. The tasks are Pre-process texts, extract terms and form concepts, and Construct hierarchy and discover non-taxonomic relations. In pre-processing texts, documents such as Portable Document Format, Extensible Markup Language are converted into ordinary texts. Stanford's POS tagger and Berkeley Parser tag words with POS and syntactic information. Multi-word terms are obtained from parsed texts to extract terms and form concepts. Articles, prepositions, descriptive adjectives, conjunctions are removed from extracted corpus. DR measure weighs each term as shown in equations (1) and (2).

$$DRM(t) = \frac{tf(t)}{max(tf)} \times \frac{|\log \lambda(t)| - \min|\log \lambda|}{\max|\log \lambda| - \min|\log \lambda|} \times \frac{df(t)}{\max(df)}, \quad \text{..................} (1)$$

Where.................. (2)
$$\lambda(t) = \frac{\max_p p^{k_1}(1-p)^{n_1-k_1} p^{k_2}(1-p)^{n_2-k_2}}{\max_{p_1,p_2} p_1^{k_1}(1-p_1)^{n_1-k_1} p_2^{k_2}(1-p_2)^{n_2-k_2}},$$

In the final task, non-taxonomic relations are discovered and hierarchy of terms is constructed. The designs used in CRCTOL are the following:
1: NP0 (including|such as) NP1 NP2 ...(and|or) ...NPn
2: NP1 is a kind of NP0, where NP0 is the hypernym of NP1 to NPn [17].
It [29] accomplishes relation abstraction (taxonomic and non-taxonomic) employing lexico-syntactic configurations and syntactic structure analysis.

Domain Ontology rapid Development Environment (DODDLE-II): DODDLE II [30] studies taxonomic relations (TR) and non-taxonomic relations (NTR). The TR are drawn from WordNet with a domain expert. The NTR are extracted from texts that are specific to a particular domain with the lexical co-occurrence statistics analysis. A few case studies in the discipline of law are carried out to evaluate the system. For TR and NTR, the precision was 30% and 59% respectively [19, 8].

HASTI: HASTI [31] as a system to retrieve concepts, non-taxonomic and taxonomic relationship between concepts, and axioms in order to build ontologies on the present kernel which is independent of any domain. So, the system will be able to build generic and domain specific ontologies from the start [19]. It is programmed for automatic ontology building. It takes Persian text as input and output is extended lexicon and ontology. HASTI uses hybrid symbolic approach as its learning methodology. Primary school text as well as story books and computer technical reports were used as case studies in which precision and recall was 97% and 88% in the former and 78% and 80% in the latter case respectively [31].

OntoCmaps: OntoCmaps [32] is independent of a domain and an OL tool that unearths deep-rooted semantic representations from a body of text. It creates abstract illustrations in the form of concept diagrams. The tool heavily depends on the internal structure of graphs to detect and extract most important elements and concepts. The authors suggested filtering mechanism based on Degree, Betweenness, PageRank and Hits metrics from graph theory. It was compared with ontology generated by Text2Onto and was observed to have returned better results [19].

OntoGain: OntoGain [33] is designed as tool for unobserved ontology acquisition (OA) from amorphous text. It retrieves taxonomic relations and does association rule mining for non-taxonomic relations by applying agglomerative clustering methods and analysis of formal concepts [34]. Multi-word domain concepts are used by this technique to fabricate a semantically strong ontology. For a comparative case study, medical and computer science domains were used for manually built as well as Text2Onto tool-built ontologies. About 70% precision for identification of taxonomic and non-taxonomic relations was obtained by the combination of agglomerative clustering and association rules in both corpora. So, it outperforms any other combination of methods [19].

ProMine: ProMine, a recent tool, encompasses consecutive application of several NLP procedures and learning processes for concept abstraction, filtration and ontology improvement. It deploys various TM and DM techniques for OL that lead to expansion and improvement of a DO rather than to improve and populate current ontology. It does three tasks: 1) extract knowledge elements from the corpus of domain documents and other sources, 2) filter concepts from the retrieved knowledge elements to find the most applicable terms of a domain and 3) categorise semantic concepts with the retrieved knowledge elements to enrich and populate DO [35].

SYNDIKATE: [36, 37] is an automated OL system that employs linguistics-based techniques to perform its OL tasks. The output of SYNDIKATE is the semantic templates and the domain knowledge that is required for the extraction of relations [8]. It combines analysis requirements from single and referentially connected body of words establishing cohesive texts. The scheme of acquiring new concepts is built on the prior knowledge of the domain.

TEXT2ONTO: Text2Onto [38] is differentiated from its predecessor, TextToOnto in three notable ways: 1) representation of the learned knowledge in the form of instantiated model primitives at a meta-level within Probabilistic Ontology Model (POM) which may then be reproduced in any expressive KR language such as OWL and RDFS, 2) facilitating interaction with the user by adding probabilities to the learned structures, 3) selectively updating the POM according to changes in the corpus by including methods that discover data-driven changes [19]. Text2Onto system extracts taxonomic associations handling Hearst pattern match technique and non-taxonomic associations handling association rule mining technique.

TextOntoEx: TextOntoEx [39] builds semantic patterns from natural domain text. Its intention is to unearth non-taxonomic associations of a particular domain type. It connects linguistic analysis with OE. To unearth candidate associations, represent their meanings and to assist in building ontology, a body of domain text is analysed. It enhances light ontology with non-taxonomic associations. TextOntoEx is useful for discovering instance of relations rather than for uncovering new relations [19]. To improve recall ratio, more semantic patterns are stored.

WEB→KB: WEB→KB is developed with a probabilistic, symbolic knowledge base that automatically creates a computer understandable worldwide knowledge base which corresponds to the content of the WWW [40]. Its methodology creates a system that can be trained to extract symbolic knowledge from hypertext, using a variety of ML methods. This system has two inputs: i) knowledge base having ontology defining the classes and (ii) training examples. The result will be: 1) instances that are classified and rules to extract new instances, 2) rules for the classification of pages and rules for recognising relations among several pages. logical and statistical algorithms are used by WEB→KB to obtain these results [8].

Semi-automatic OL Tools

Acquisition of Semantic Knowledge Using Machine Learning Methods (ASIUM): ASIUM's [41, 42] objective is to assist an expert to acquire semantic knowledge and taxonomic relations from concepts that are retrieved from technical texts by making use of syntactic analysis. It is done by associating the frequency with which each word in a given text occurs using conceptual and hierarchical clustering techniques [15]. French texts are given as input. A metric is used in computing semantic resemblance amongst clusters. Conceptual clustering method is used to successively group clusters in order to form concepts of ontology. Two-step formula is used to accomplish its results: viz., factorization (conceptualization) and clustering (ontology building). The technique of pyramidal clustering is adopted to construct ontology level by level.

Caméléon: Caméléon [43] is programmed to enrich conceptual models by learning conceptual relations. It depends on linguistic principles for relation recognition. Some prototypes indicate the same relation type while others disclose domain-specific associations. Learning conceptual relations has a twofold procedure. The first, identifies pertinent patterns and associations. The second, using those patterns identifies lexical relations and enriches a conceptual model. Patterns help in listing potential lexical associations in the texts. Ontology evaluation improves conceptual relations.

Language Technology Group (LTG) Text Processing Workbench: The LTG [44] is composed of a group of computational systems to disclose inner structures in NL texts scripted in English language. In LTG, OL is done in two successive phases viz., representation and analysis. In the first phase, the script is changed to elements of interest. In the second phase, important correlations in the texts are found using tools for statistics gathering and inference to analyse the features. The tools for analysis work on an abstract level of the feature elements which are represented as SGML items. So, the tools are not dependent on any specific supposition about the character of the feature set [9]. For lexicographic and statistical language modelling purposes, the workbench is used. The methods used are taken from IR, computational linguistics, and knowledge engineering (KE).

Mo'K Workbench: Mo'K Workbench [45] is a configurable workbench developed by INRIA, LRI Univ. Paris-South [46]. It helps semi-automatic building of ontologies using diverse conceptual clustering methods. It helps the expansion, comparison, description and evaluation of various conceptual clustering methods. It also provides precise definitions for the measurement of similarity and operators used for the construction of classes. Thus, they ease method instantiation and configuration tasks [9]. Corpus as input is taken for learning process by this workbench. With the help of NLP techniques, a list of triplets is obtained from the body of text. A verb, a word with its syntactic role in a sentence forms a triplet. Mo'K computes the frequency of occurrences of each one. The triplets with low and high frequency of occurrences are deleted from list. Lastly, Mo'K can help in the formation conceptual clusters by calculating the semantic distance between the triplets in the previous list [9].

OntoLearn: OntoLearn [47, 48, 49, 50] extracts related domain terms, associates them to apt concepts, and finds relations among the concepts. It uses linguistics and statistics-based techniques like NL analysis and ML techniques to perform its OL tasks. Tasks are as follow: Pre-process texts and retrieve terms, form concepts, and construct hierarchy.

In the first task part-of-speech (POS) tagging and sentence parsing tools are used to pre-process the text. Relevance analysis assumes two metrics: domain relevance (DR) and domain consensus (DC). DR calculates the specificity of a term t in comparison to the target domain Dk by comparatively analysing a list of predefined domains D1, . . . , Dn. The measure is defined as in the equation (3)

$$DR(t, D_k) = \frac{P(t|D_k)}{\sum_{i=1...n} P(t|D_i)},$$ ……………… (3)

where $P(t|D_k)$ and $P(t|D_i)$ are estimated as $\frac{f_{t,k}}{\sum_{i\in D_k} f_{t,k}}$ and $\frac{f_{t,i}}{\sum_{i\in D_i} f_{t,i}}$, respectively. $f_{t,k}$ and

ft,i are the frequencies of term t in domain Dk and Di , respectively. DC gives the number of times a terms appears in a single document in contrast to its overall appearance in the target domain. The entropy of the domain consensus of a term t in domain Dk is an defined as equation (4)

……………… (4) $$DC(t, D_k) = \sum_{d \in D_k} P(t|d) log \frac{1}{P(t|d)},$$

where P(t|d) is the probability of coming across term t in document d of domain Dk [17].
Form concepts use concepts and glossary from WordNet to select the best sense combinations. Construct hierarchy discovers hypernyms from WordNet to arrange concepts in domain concept trees.

Prométhée: Prométhée [51, 52] extracts and refines lexical-syntactic patterns associated with concept relations in technical corpora. It enriches pattern bases extracted during learning. To improve patterns, Eagle learning system [53] is proposed. The logic behind this system is inductive paradigm learning using examples [54]. It obtains intentional descriptions of concepts from their extensional descriptions.   The acquired descriptions are subsequently utilized for identification as well as for classification tasks. After Prométhée has retrieved lexical-syntactic patterns, some events of the patterns are extracted from the corpus and placed as examples of the pattern and Eagle produces descriptions that are taken as limitations from these labelled patterns refining the patterns [15].

Semi-Automatic Domain Ontology Acquisition Tool (SOAT): The SOAT [55] permits semi-automatic domain ontology (DO) acquisition from a domain corpus. Its primary aim is to obtain associations from parsed sentences applying phrase rules to recognize main words with strong semantic connections like hyperonyms or synonyms. InfoMap [56], a KR framework, is used for acquisition process. It is constructed to accomplish NL comprehension and catch the theme words, e.g.: pairs - one or two nouns with a verb, in a stream of text with a delimiter. Taxonomic and non-taxonomic are two key forms of relations among concepts in InfoMap. Domain corpus with POS tagging is given as input to the tool. Usually a keyword in the domain is chosen to be a root in the corpus. Then the procedure, by deploying extraction rules, targets to look for a new keyword apparently connected to the previous one. Implementing the guidelines and the structure fixed in InfoMap, it adds the new keyword to the collection of ontology. The new keyword is used as a root for an iterative process of a specified number or till there is no possibility of finding a new related keyword [15]. SOAT enforces that corpus quality to be high.

SubWordNet Engineering Process Tool: WordNets follow iterative SubWordNet Engineering process approach [57]. SubWordNet is built on WordNet semantic structure. It includes the capacity for discovering concept elements, identifying concept and maintaining them. Three modularised layers, the graphical user interface (GUI) layer, the process layer, and the data layer, are used to perform each of these functions [9].

TERMINAE: TERMINAE [58, 59] integrates tools associated with linguistic and KE. Taking into account its corpus and analysis of frequency of term occurrences, the linguistic tool permits naming terminological forms.

Ontologists study the term usage in a corpus to label denoted term meanings. KE tool contains an editor and a browser. Terminological forms are represented as terminological concept in TERMINAE.

Text-To-Onto: Text-To-Onto [60] has a structure for semi-automatic OL from texts that employs a range of algorithms for various OL subtasks. Its framework uncovers abstract theoretical structures and engineering ontologies.   Three processes that make Text-To-Onto unique for taxonomic relation extraction are: 1) the agglomerative clustering, 2) WordNet hypernyms and 3) lexico-syntactic patterns. For, non-taxonomic relations extraction association rule mining is used [61]. Iterative specialisation of a generic ontology or WordNet is suggested. Automatic learning using examples of patterns and association rules help in the discovery of concepts and relations between concepts. After the ontology is pruned using statistical measures, it is presented to the expert for evaluation [62]. To develop and maintain ontology tasks, DM and NLP techniques are used. It is integrated into KAON system. It has a collection of multi-strategy learning approaches for different inputs and tasks. The advantage is varied algorithms help term extraction, taxonomy construction and learning relations among concepts [8, 19].

TextStorm and Clouds: TextStorm/Clouds [63, 64, 19] is composed of TextStorm and Clouds to accomplish complementary activities in building a semantic network.   As semi-automated OL system it aims to construct and improve DO for use in Dr. Divago to discover musical pieces or drawings in a multi-domain environment [17]. It follows linguistics-based techniques and logic to perform OL tasks such as: a) Pre-process texts and extract terms, b) Construct hierarchy, discover and label non-taxonomic relations and c) Extract axioms. In TextStorm/Clouds, parts of speech tagging, syntactic structure analysis, WordNet, and anaphora resolution are used for extraction of both taxonomic and non-taxonomic relations [63]. TextStorm, as NLP tool is used to retrieve binary predicates from a text using syntactic and discourse knowledge by creating a list that is sent to the Clouds tool. The construction of an interactive semantic network is done by Clouds [15].

Word Learning From Interpreted Examples (WOLFIE): WOLFIE [65] studies and acquires semantic lexicon from a body of text. The acquired lexicon contains words juxtaposed with meanings represented. WOLFIE permits synonymy and polysemy. It in an integral part of a system which combines features like, handling of polysemy and synonymy; interact with CHILL [66] and lastly trades for the finest assortment of phrase meanings founded on several heuristics.   The algorithm is based on the premise that the choice of the lexical item will inhibit the possible meanings of terms that are not learned. Certain assumptions about the problems are made in order to achieve the goal of the algorithm: possible meanings of words and phrases in a sentence gives the meaning of that sentence; no noise can be present in the sentence representation; and the meaning of each word occurring in a sentence is found only once in its representation. [9]

LexOnt: [67] is partially-automatic ontology building system whose resources are Programmable Web directory of services, WordNet, Wikipedia, and the existing ontologies to unearth related expressions. Ontologies are constructed by iteratively interacting with the user. Terms can be added to the ontology and the terms can be ranked by the user. LexOnt is a plugin for Protégé ontology editor that facilitate the ontology creation process by interacting with the user. Unstructured text is sent as input to the system [19].

OntoGen: OntoGen [68] supports in the construction of ontology by unearthing probable concepts and associations in a body of texts by means of ML and TM algorithms. The tool adapts monitored approaches for concept discovery. The system employs enhanced user graphical interface to select a topic which in turn indicates certain probable subtopics from a list of designated articles. The obtained concepts are authenticated by the user.

Non-automatic OL Tool

TFIDF-based term classification system: TFIDF [70] aims to find terms to learn associations that bind them in a domain. The tool operates with three components, viz., single-word term classifier, lexico-syntactical pattern

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finder and relation extractor. The first extracts lone words in a domain. The second has two sub-modules: the first sub-module learns patterns among known relations and applies interfaces to inter-operate between two systems; and the second sub-module learns patterns on term co-location methods. The relation extractor is the last component. The input is an annotated domain corpus, parsed using a shallow NLP tool.

Research Issues

OE is powered by active research in the area of OL by various methods and systems. Thirty OL tools have been surveyed as part of the study. The study reveals that in recent years not many OL tools have being reported. Of the available tools, numerous techniques like, KA, ML, IR, NLP, AI, etc are implemented. The research issues are grouped under Combining approach, Human intervention and Evaluation method.

Combining Approach: Not many tools have an amalgamation of techniques or incorporating new ones [71]. Tools have dearth of frameworks for a mix of comparison of various extraction methods [19]. ASIUM, KAON-TextToOnto, Ontolearn, OntoLT, DODDLE II, and WEB->KB combine certain linguistic analysis procedures along with ML algorithms in order to unearth conceivable concepts and associations among them. Tools and structures for KA, NLP and Knowledge Management are autonomous from each other, even though a number of components are shared between them. Currently, a very little work is reported on tool integration. Reinberg and Spyns indicated that there has been no publication of a comparative study of the efficiency and effectiveness of different techniques applied to ontology learning [72]. It is also noted that no single method could be efficient by itself without a combination of techniques depending on the application in use [21]. Aussenac-Gilles suggested that one of the research challenge of SW is to make a list containing existing techniques, their attributes and possible amalgamations that would be a useful pointer to progress towards a tool or a technique combination leading to specific processes [73].

Human Intervention: Linguistic and statistic approaches do have shortcomings. Therefore, as of now, human intervention is inevitable to augment better outcomes, that is to say, full automation of OL may not be an immediate reality [19].As for semi-structured methods, it can be seen that recent work has been done providing better pre-processed input such as better tagging in [74]. Recently work on creating different language WordNets have been seen such as Romanian [75], Japanese [76], Chinese [77], Persian [78, 79] and Arabic [80] to name just a few. The growth of such WordNets will help OL systems which use the Hjelm's multilingual view to create better ontologies.

Evaluation Method: A noteworthy challenge/open problem for ontology creation/learning/development from text is lack of ontological gold standards for OE [19, 21]. According to Gacitua, Ricardo, et al., for evaluating the effectiveness of ontologies, the techniques giving optimal performances for the ontology process have to be determined which is not a trivial task [71]. Existing OL tools are not designed to measure the exactness of learning process, and not compare results obtained from various learning techniques. Hence, the final ontology is ought to be evaluated by an ontologist [15]. Recent applications use plain text and semi-structured text as their inputs to improve their OL results as seen in ISOLDE, GALEON, and OntoCmaps. Also text based applications are working on combining different OL methods improve their results [81]. To be able to better evaluate these results a few of them have chosen to create frameworks to provide an environment to test different methods and compare their results such as in [81, 82, 83, 84].

CONCLUSION

OL systems aid KA process in ontology construction. Yet, the ambition of automating OD is far from reality. The review shows there is a little conformity amongst various OL approaches and systems developed. These applications provide partial technological aid to process steps suggested in diverse approaches excluding in Kietz et al's and Text-To-Onto system [1]. A tool for complete automation of OL process is a distant reality. However,

some current tools focus on gaining lexical-semantic knowledge while some others aid in extracting concepts or associations from a cleaned body of text with the help of the user [1]. The available tools represent only a few steps of the entire process and development of an ontology, not being part of a larger methodology for the development of ontologies. Most methods for OL from texts are based on NLP techniques and statistical measures. All these methods require the participation of a knowledge engineer and a domain expert to evaluate the final ontology and also the progress of the process. An evaluation of ontologies is a complex topic in itself and is usually measured by the ontology's ability to respond to a set of questions which it is supposed to bring answers to. Although it is an area that still needs a lot of advancement, the use of these tools helps the actors involved in the construction/enrichment of ontologies, reducing the development time and cost by extracting knowledge from the texts. However, in the current stage of development of these techniques, it will always be up to human actors to arrive at a final decision to incorporate a concept or to make some relationship between concepts in the ontology.

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RESEARCH ARTICLE

Variable Step Incremental Conductance MPPT for the Boost Converter in Grid Connected Solar PV System

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ABSTRACT

In this paper, a variable step incremental conductance maximum power point tracking (MPPT) algorithm is proposed to extract maximum power from a photovoltaic (PV) system. The Power (P) –Voltage (V) and Current (I) – Voltage (V) characteristics of the PV array are presented for different irradiance levels and cell temperatures. The accuracy and tracking speed of the system are improved with this algorithm. Simulation work is carried out in MATLAB/Simulink environment. Simulation results show a better response of the grid-connected solar PV system under rapidly changing environmental conditions.

Keywords: Boost converter, Grid-connected photovoltaic (PV) system, Maximum power point tracking (MPPT), Voltage Source Inverter (VSI), Grid voltage.

INTRODUCTION

Solar PV energy is a promising field of renewable energy sources due to its distributed nature, free of cost, and pollution-free [1]. By implementing the proper methodology, maximum energy can be harnessed from the sun. PV technology is used for small and large-scale power generation in all countries. The PV technology is used in two combinations such as grid-connected and standalone systems. Solar energy is directly converted into electrical energy through the Photovoltaic effect [1-3]. However, the conversion efficiency of a solar cell is less. For standalone PV systems, a battery storage system is required. Renewable energy resources are being green and non-pollutant, and meet the needs of the energy delivery of the developing population. Due to globalization and industrialization, the power requirement is growing rapidly. By reinforcing new technology in the renewable power field, maximum power from solar may be obtained with the minimal loss [1-2]. Because of smooth maintenance, being extensively dispensed, and pollutants free, PV is the most promising technology [3, 4]. In this system, PV panels, DC-DC

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converter with MPPT, and DC-AC converter are required to combine PV with the grid. MPPT is a technique that maximizes the extraction of power from PV under any advert condition. Fractional open-circuit voltage (FOCV), fuzzy logic (FL) fractional short-circuit current (FSCC), particle swarm optimization (PSO), perturb and observe (P&O), artificial neural networks (ANN) and incremental conductance (INC) are the different types of MPPT techniques used for solar PV systems [1-15]. Variable step INC MPPT is an improved technique to increase the accuracy of the MPPT [1]. In this paper, grid-connected PV with variable step INC MPPT is discussed with relevant results and it is organized as follows: Section 1 deals with the basics solar cell and its characteristics under dynamic climatic conditions. The proposed MPPT algorithm is demonstrated in section 2. Simulation results and discussions are presented in section 3. Conclusions are given in the last section.

PHOTOVOLTAIC CELL

A single PV cell is capable of converting solar energy into electrical energy directly. The typical equivalent circuit of a single diode model is depicted in Figure1. It is the basic element in a PV array with low conversion efficiency. P-V and I-V characteristics of PV array with MPP are shown in Figure 2. In any PV module, the Impp is lower than the short circuit current Isc and the Vmpp is lower than the open-circuit voltage Voc. Number solar cells form a module and collection of such modules constitute a PV array. In a PV array, solar cells are in series-parallel connections.

Mathematical equations derived from the equivalent circuit are written as

$$I = I_{pv} - I_d - I_{sh} \tag{1}$$

$$I = I_{pv} - I_s \{ exp[\frac{q(V+IR_s)}{mkT}] - 1 \} - (V + IR_s)/R_{sh} \tag{2}$$

Where I_s is the Diode saturation current(A), I_{pv} is Photocurrent of the PV cell(A), I_d is diode current(A), I_{sh} - shunt resistor current(A),m- diode quality factor, q- Charge of an electron(1.6×10⁻¹⁹ C),I-output current(A), R_s - Series resistance(Ω)R_{sh} - shunt resistance(Ω),T- Temperature(K)and k- Boltzmann constant (1.38×10⁻²³ J/K). The maximum current and power are expressed as

$$I_{mp} = I_{pv} - I_s \{ exp[\frac{q(V_{mp}+R_sI_{mp})}{mkT}] - 1 \} - (V_{mp} + I_{mp}R_s)/R_{sh} \tag{3}$$

$$Pmax = V_{mp} \{ I_{pv} - I_s \{ exp[\frac{q(V_{mp}+R_sI_{mp})}{mkT}] - 1 \} - (V_{mp} + I_{mp}R_s)/R_{sh} \} \tag{4}$$

Figure 3 shows the characteristics of a PV array at different temperatures with a constant 1000W/m² irradiance level. It is observed that power produced will decrease with an increase in temperature. The change in the cell temperature severely affects the performance of the system as shown in Table1. Figure 4 shows the performance of the PV array at different irradiance levels. When the solar irradiance is varied from 250- 1000W/m², the generated power is increasing in the array and reaches the maximum at 1000W/m².

PROPOSED VARIABLE STEP INC MPPT ALGORITHM

Figure 5 shows the typical P-V characteristics of a PV array. The point MPP is the peak point at which PV power is maximum. The proposed variable step incremental conductance algorithm target this unique point with good accuracy. The proposed algorithm shown in Figure 6 is a modified form of conventional INC MPPT in which the step size is kept optimal to get better accuracy without compromising the tracking speed. In the algorithm, the PV array conductance (I/V) is compared with the incremental conductance of the PV array (dI/dV). The flow chart in Figure 6 is used to explain the flow of the algorithm. Equations related to this algorithm are expressed as follows.

$$dP/dV = d(VI)/dV = IdV/dV + VdI/dV \tag{5}$$

$$dP/dV = I + VdI/dV \tag{6}$$

$$(1/V)dP/dV = (I/V) + dI/dV. \tag{7}$$

$$dP/dV = 0, \text{ at MPP} \tag{8}$$

$$dP/dV > 0, \text{ Left of MPP} \tag{9}$$

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dP/ dV< 0, Right of MPP   (10)
dI/ dV   = − I /V, at MPP   (11)
dI /dV> − I /V, Left of MPP   (12)
dI /dV< − I /V, Right of MPP   (13)

Where dP is the variation in output power of PV array, dV is the variation in output voltage of PV array and dI is the change in PV current.

D (n) = D (n-1)± d   (14)

$d = N * \left| \dfrac{dP}{dV} \right|$ (15)

Where, D(n) is Duty cycle, d is the variable step size and N is the scaling factor. The step size selection is based on dP/ dV ratio and the value of N decides the performance of the proposed MPPT. If the tracking steps are small, accuracy increases but tracking speed decreases. If the step size is large, accuracy decreases. However, dP/ dV ratio is the key parameter used to increase or decrease the step size of the proposed algorithm. In this work, simulation of the proposed algorithm is carried out with step size d=0.2 and scaling factor N=0.15.

SIMULATION AND RESULTS

Simulation of the grid-connected PV system is carried out with MATLAB/Simulink software. Signal builder block is used for setting up the irradiance and temperature profiles. Simulink model of the grid-connected PV system is shown in Figure 7. The simulation parameters required for this system are shown in Table2. Simulation results are presented with respect to time (sec) at different irradiance levels (250-1000) W/m^2 and different temperatures 25-50^{0}C. Figure 6   shows the flow chart of the proposed MPPT algorithm with variable step size and the simulation parameters are listed in Table2. Irradiance and temperature profiles used for the simulation are shown in Figure 8. With a small modification in conventional INC MPPT, the tracking speed of MPPT is enhanced.

CONCLUSION

In this paper, a simulation of a grid-connected PV system with improved performance is presented and it is suitable for small-scale industrial and residential applications. The proposed MPPT improves the accuracy of the MPP tracking under rapidly changing irradiance and temperature levels. The variable step size is used in the incremental conductance method to improve the tracking speed and accuracy. Improvement in the overall system performance is made possible with a simple and efficient algorithm. Simulation results have confirmed the better performance of the system with the variable step INC MPPT.

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Table1. Effect of temperature on Power at MPPT

Temperature ^{0}C	Pmpp (W)
25	1000
35	950
50	900

Table 2. Simulation Parametersfor 100kW system

Parameter	Value
Open Circuit Voltage (Voc)	64.2 V
Maximum Power Voltage (Vm)	54.7 V
Short Circuit Current (Isc)	5.96 A
Maximum Power (Pm)	305.2 W
Diode saturation current (Is)	1.1753e–08
Diode ideality factor	0.93246
Shunt Resistance (Rsh)	993.51 Ω
Maximum Power Current (Im)	5.58 A
Series Resistance (Rs)	0.0379 Ω

Uma Maheswari
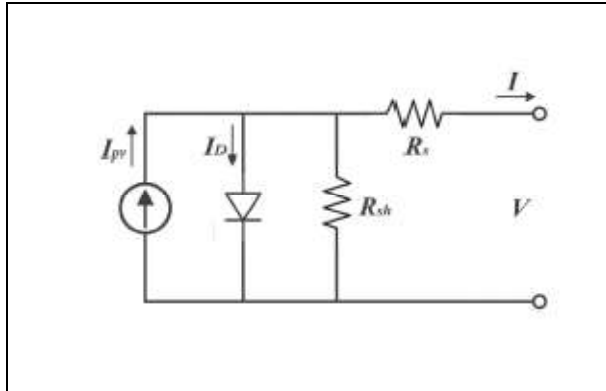
Figure1. Equivalent circuit model of a PV Cell
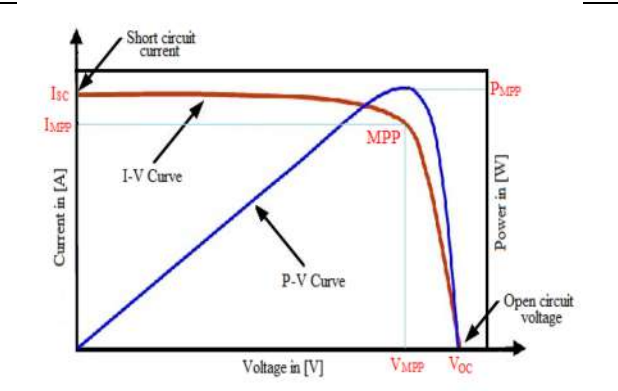
Figure 2. I-V and P-V characteristics of PV module with MPP
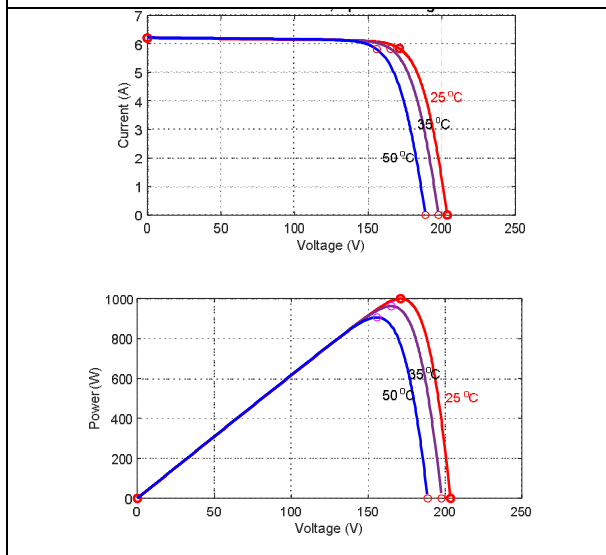
Figure.3 I-V and P-V characteristics at different temperatures
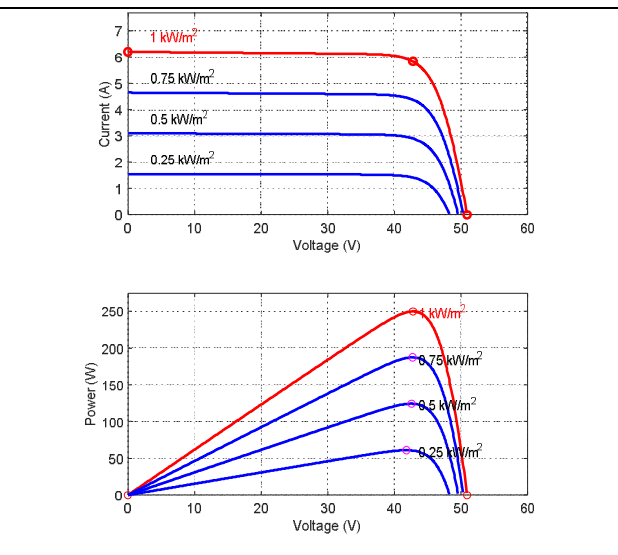
Figure 4.I-V and P-V characteristics at different irradiances
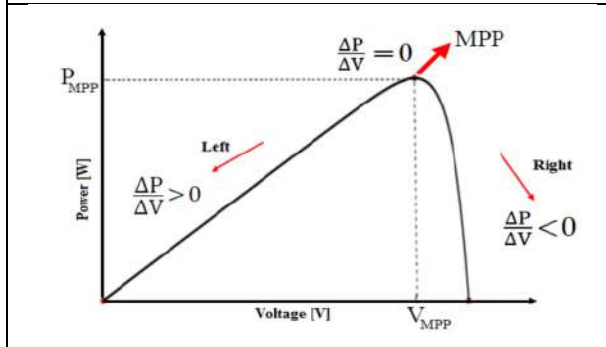
Figure 5. P-V characteristics of PV array
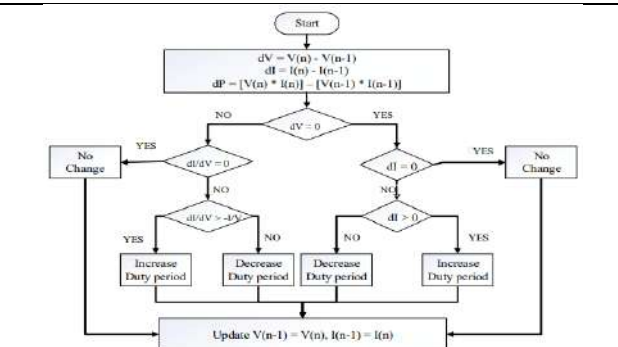
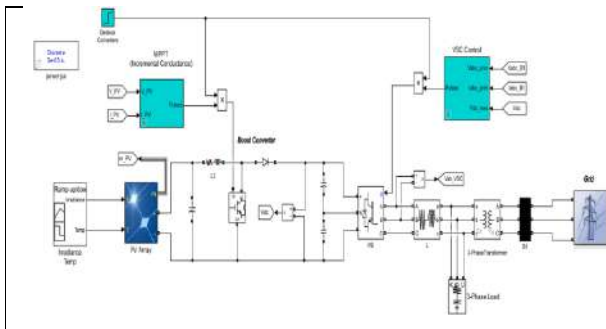**Figure 6. Variable step incremental conductance MPPT Flowchart**

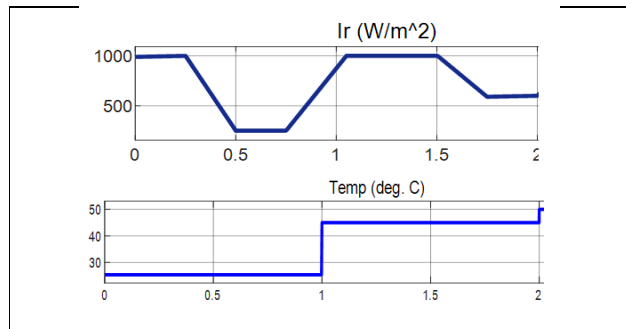**Figure7. Simulink model of Grid connected PV system with MPPT**

Figure 8. Irradiance and temperature profile with respect to time in seconds
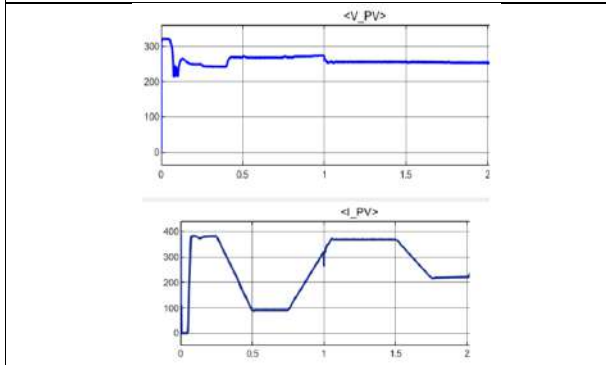
Figure 9. PV array voltage (V) and current (A)
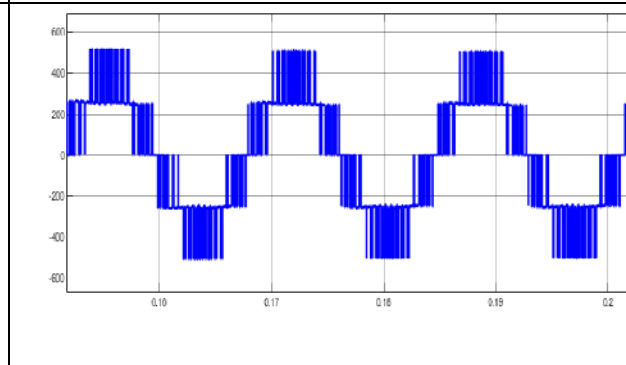
Figure10. InverterOutput Voltage (V)
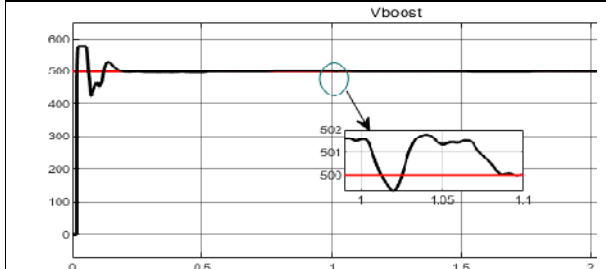
Figure11.Boost converter output voltage (V)
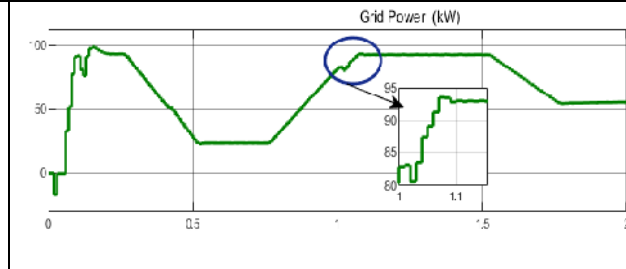
Figure 12. Grid power (kW)
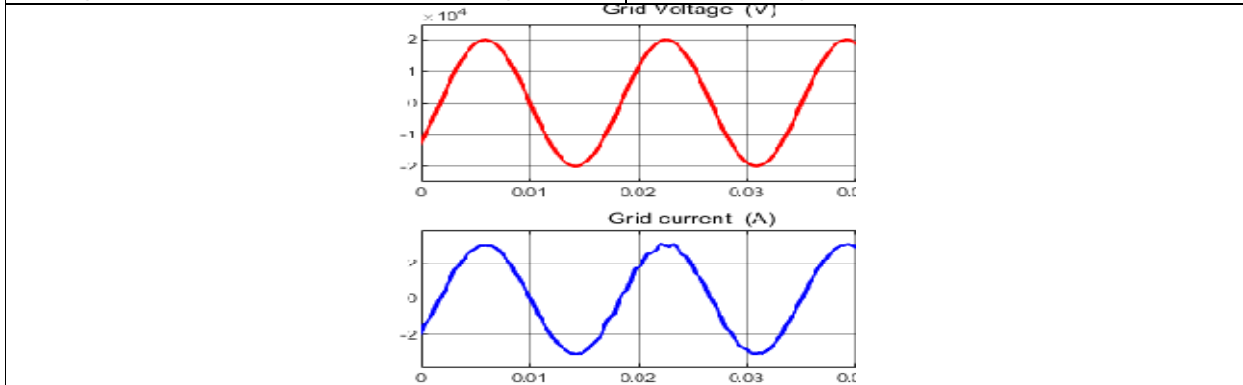
Figure 13.Grid voltage (V) and current (A)

RESEARCH ARTICLE

Impact of an Artificial Intelligence in Natural Language Interface to Database

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ABSTRACT

The diverse business needs and smart technological advancements require the simulation of human activities to be accomplished through computing machines efficiently. Techniques employed for Information retrieval from natural languages are still flourishing area of research. The goal of the work is to create efficient artificial intelligent model to recognize natural language text written in Kannada from the user and convert it into English statement and then generating the SQL Query. Finally output the solution to SQL Query from the database.

Keywords: Natural language processing; database; Artificial intelligence.

INTRODUCTION

The information retrieval system is deals with organizing heterogeneous information stored in digitized form. Recognizing character information present in digital document is the work which comes under pattern recognition. Optical Character Recognition (OCR) is one of similar tool used to interpretation of contents present in the document using computing machines. Early works focused on development of OCR tool for Roman and Latin language scripts. Further, the progressions towards colorful Indian language scripts are accorded to extend the functionality of OCR to read numerous scripts. However there being numerous successful examinations on OCR towards Indian scripts, there are still numerous grueling exploration issues to be addressed in this regard. One of the critical walls that hamper the path to reach the successful recognition rate is the complexity involved in script. Especially, this is true with numerous of the South Indian Language (SIL) scripts [1] due to their wide character set and structural diacritics. Thus, it's essential to address the complications of SIL scripts in order to reach advanced recognition rates. This work focused on analysis of involvement of artificial intelligent technique with various classification models [2] in Kannada sentence recognition. Translator module used to translate characters into ASCII code. AI technique based

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query interface helps user to interact with system in more expressive natural language. Here database systems integrated with SQL to define its schema. Section 2 provides related work. Details on the created datasets are described in section 3. Recognition of Kannada character using deep learning integrated classification models and conversion of database to SQL Query is presented section 4. Section 5 deals with conclusion.

LITERATURE SURVEY

K. Javubar et al. [3] paper discusses that the information extracted works on user-based natural language queries on the health domain. Recently, a significant work is proposed in the context to Indian languages and script. Most of the character recognition work is based on Devanagari Script. Devanagari numeral recognition using zonal features presented in K. Johnson et al. [4]. I.K. Sethi et al. [5] presented method on hand-printed Devanagari numerals that uses binary decision tree classifier. Three different features like density, moment and descriptive component are used for classification of Devanagari Numerals proposed by R. Bajaj et al. [6]. B. K. Verma [7] used Multi- Layer Perceptron (MLP) networks and the Radial Basis Function (RBF) in recognition of handwritten Hindi Character. M. Hanmandlu et al. [8] proposed a Fuzzy model to recognize the handwritten Hindi numerals and characters. In N. Sharma et al. [9], the handwritten Devanagari characters are recognized using chain code features and quadratic classifier. In G.Navarro [10], the character encoding and regular expressions are used for recognition. A. Goyal et al. [11] designed OCR using pre-processing, feature extraction technique and classification algorithms. In S. Kumar [12], five feature-extraction methods are compared and tabulated for handwritten characters. In K.V. Kale et al. [13], the Central moments, invariant moments and Zernike moments are used for recognition of characters of Marathi languages are reported. The Devanagari numerals are recognized using shape descriptors like Higher order Zernike moments for classifying in G. Amayeh et al.[14]. The invariant moment features are used for Marathi Handwritten numerals recognition presented in G.G. Rajput et al. [15]. An efficient Devanagari character classification system was proposed using SVM in S. Puri et al. [16]. The model is used to recognize characters of Hindi, Sanskrit and Marathi text images. M.A. Rahman et al.[17] proposed the modified syntactic approach for classification

A review of the research related to handwritten character recognition of Indian regional scripts is presented by U. Pal et al.[18].Recognition system for South Indian scripts are detailed in M. A. Rahiman et al.[19].Various approaches used for recognition of Online Handwritten Malayalam Characters are proposed by A.M. Chacko et al.[20]. Support vector machine is used for recognition in Tejo et al. [21]. Structural feature and Feed forward back propagation network are used for recognition of Malayalam character in J. John et al.[22]. The discrete features are used for recognition of handwritten Malayalam characters in K. P. Primekumar et al. [23]. Comparison of Hidden Morkov Model with SVM classifier is presented. In E. Kavallieratou et al. [24], the given image is divided into different zones and features are extracted from each zone and Multilayer Perceptron is used for classification. Extended zonal representation to improved character classification in Malayalam is proposed in P.V. Raveena et al. [25].Scattering network model for classification of Malayalam characters is presented in K. Manjusha et al. [26]. Improvement in recognition is observed compared to pixel level feature extraction method using Softmax regression classifier. Although, the analysis on SIL scripts conducted is appreciable, the features selected using various extraction methods, which gives additional burden. So there is a need of techniques for automatic feature extraction before classification task.

PROPOSED FRAMEWORK
Creation of database
Any newly structured model must be empirically tested for different qualities so as to evaluate its viability and furthermore its proficiency to satisfy the requirements. To this end, broad experimentation must be led on a relatively enormous dataset. In this view, an effective attempt is made to create an unconstrained archive datasets written in Kannada contents. Database will help researchers to implement and compare the performance. The first task of the work was to build a corpus for experiment. The Visual analysis of printed characters of Kannada character is curved and symmetry in nature. The handwritten shapes do not exhibit perfect symmetry as compared to printed characters. The small circles, knots decrease the recognition rate in the presence of noise. While using

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some Image Processing algorithms used to bring images to proper form. Database is standardized by size ranging from 32×32 to 500×500. Resizing of image below 32×32 leads to loosing of some textual information. Further image quantity is increased by using augmentation tactic.

Processing and Recognition of character
Kannada character images are preprocessed and deep learning with features from convolution neural network is considered for classification. Convolution Neural networks (CNN) are variants of multi-layer perceptron with spatial local correlation in natural images are exploited well. These are bonded with a feed forward neural network to form Deep convolution neural networks (DCNN). It consists of differing kinds of layers, convolution and pooling generally alternated as shown in the figure 1. Convolution filters are used in initial layers, where the depth of every filter will increase from left to right within the network. The last stage is often manufactured from one or more totally connected dense neural networks layers. These Fully connected (FC) layers are generally connects to the output layer and constructs the desired number of outputs where "Softmax" activation function is used. For input and intermediate layers, Rectified Linear Unit (RELU) activation function is used. Gradient will not be changed in this activation function. So it will not back-propagate any errors.

The proposed Deep convolution Neural Network (DCNN) model is based on famous very deep convolution networks called VGGNet architecture. Kannada character image is smaller in size compared to ImageNET database objects, so input size is reduced and standardized to 32X32 and given to the model. To classify the characters, three fully connected layers of classifier model replaced by 2 fully connected dense layers, a flatten layer and dropout layer with drop out ratio of 0.6. By training, the weights of the layers are adjusted to recognize proper characters. Transfer learning based approach is adopted here where basic VGGNet deep learning model is trained on ImageNet dataset. Stochastic gradient descent method is used to learn the model with Soft max activation function which is given in the equation (1).

$$\sigma(z)_j = \frac{e^{zj}}{\sum_{k=1}^{K} e^{z}k} \, for \, j = 1,2......k \qquad (1)$$

Back propagation model with a momentum of 0.1and learning rate of 0.01 is applied. Architectural details of proposed model are given in the table 1.

The Design of this system can be explained as a combination of Natural Language Processing and Machine Learning. In Deep convolution neural network model, features are extracted from input image in the intermediate layers of network. Then it is passed to output layer of DCNN model which has Softmax classifier. These features are also given to popular machine learning classifiers such as k-NN classifier, Naive Bayes, SVM, PNN classifiers. Classification results are compared with Random forest classifiers, which is ensemble classifier. It has set of decision trees to accurate classification and recognition of characters which together form English sentence.

IMPLEMENTATION
A total of 90000 characters belonging to basic character corpus are considered as database. Train, validation sets are taken in a proportion of 80:20. That means 72000 training images, 18000 validation images. All images in the dataset are resized to 32X32 pixels. Deep learning model requires large training data to get accurate results. So the number of images in each character class is increased to 2000 using augmentation approach. Input image is given to the DCNN model to extract features and finally classification results proposed model are compared with other shallow learning methods.

Evaluation

For the customized database, learning process of DCNN model is shown in the Figure 2. It shows an accuracy of 93.01% on the validation dataset after 14 epochs. Accuracy measures of various classifiers with CNN features are shown the table 1.

Conversion of Natural Language Query to SQL Query

After recognition of Kannada scripts, Artificial intelligence integrated translator tool is used to convert text into English sentence. Translated data is fed into the Artificial Neural Network model for the conversion of Natural Language Query to SQL Query. Tokenization is breaking up a sequence of strings in to smaller units such as words, keywords, phrases, symbols and other elements called tokens. For example, if the given input query is "find the employee salary", then in here each word of the sentence, i.e. find, the, employee, salary will be stored in a list like ['find', 'the', 'employee',' salary']. The stop word filtered input is taken and is combined with Database and Column names of the database. The column names as well as the database names are given as the input so that artificial neural network model, which understands that a word in the input is of the particular column and a database. It is quite essential part of the system for the artificial neural Network to identify the column and database name given in the input. The model has been trained on select, count, average, minimum, maximum, sum, AND operation, OR operation, between operation, order by, where, group by- these commands are available during the conversion of SQL Query. Processing query will generate necessary result as in figure 3.Perfomance analysis shows 92% accuracy for properly generated query result as is figure 4.

CONCLUSION

An efficient manner to recognize characters using combination of CNN feature extractor with different classifier is proposed. The experiment has been conducted on Kannada characters. This study presents an idea for converting a Kannada text into a SQL query. This is accomplished by first turning the sentence into English text, then constructing a SQL query. The data is taken from the database when the query has been formed. The goal of the work is to build data management process accessible to common people. To access databases, the user does not need to master a sophisticated query language like SQL.

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Table 1 Accuracy and score time for different classifiers

Sl.No	Method	Accuracy in %	Time taken for prediction in seconds
1	DCNN	93.01	64.0
2	CNN-k-NN	81.9	86.3
3	CNN-Naïve Bayes	58.9	74.02
4	CNN-Random Forest	72.93	55.97
5	CNN-SVM	67.90	69.37
6	CNN-PNN	82.17	81.5
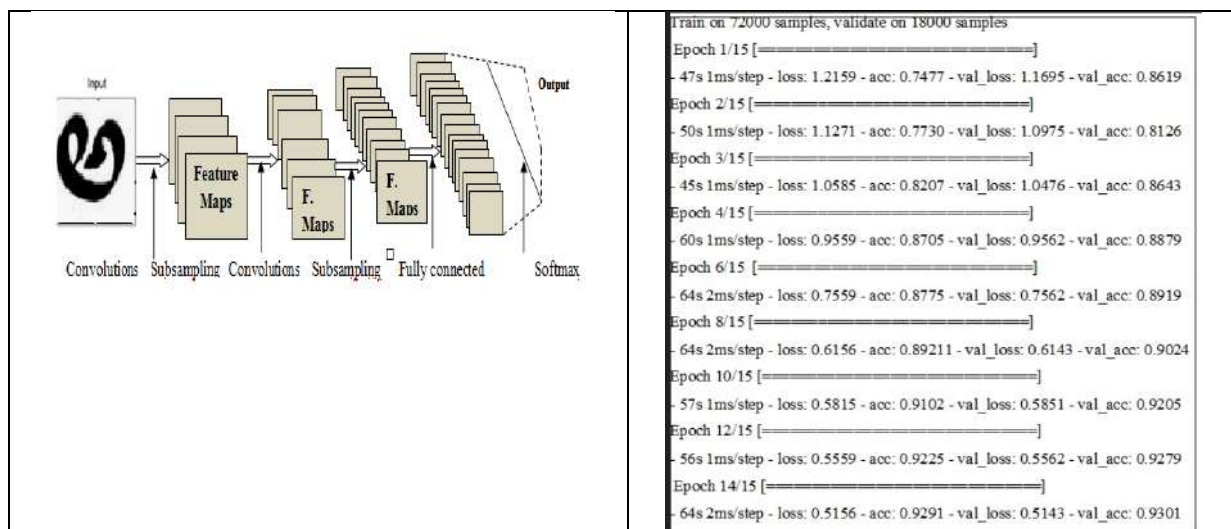
Fig 1. Deep Convolution Neural Network Model (DCNN)

Train on 72000 samples, validate on 18000 samples

Epoch 1/15 [=====================]
- 47s 1ms/step - loss: 1.2159 - acc: 0.7477 - val_loss: 1.1695 - val_acc: 0.8619

Epoch 2/15 [=====================]
- 50s 1ms/step - loss: 1.1271 - acc: 0.7730 - val_loss: 1.0975 - val_acc: 0.8126

Epoch 3/15 [=====================]
- 45s 1ms/step - loss: 1.0585 - acc: 0.8207 - val_loss: 1.0476 - val_acc: 0.8643

Epoch 4/15 [=====================]
- 60s 1ms/step - loss: 0.9559 - acc: 0.8705 - val_loss: 0.9562 - val_acc: 0.8879

Epoch 6/15 [=====================]
- 64s 2ms/step - loss: 0.7559 - acc: 0.8775 - val_loss: 0.7562 - val_acc: 0.8919

Epoch 8/15 [=====================]
- 64s 2ms/step - loss: 0.6156 - acc: 0.89211 - val_loss: 0.6143 - val_acc: 0.9024

Epoch 10/15 [=====================]
- 57s 1ms/step - loss: 0.5815 - acc: 0.9102 - val_loss: 0.5851 - val_acc: 0.9205

Epoch 12/15 [=====================]
- 56s 1ms/step - loss: 0.5559 - acc: 0.9225 - val_loss: 0.5562 - val_acc: 0.9279

Epoch 14/15 [=====================]
- 64s 2ms/step - loss: 0.5156 - acc: 0.9291 - val_loss: 0.5143 - val_acc: 0.9301

Fig. 2 Training and validation accuracy values
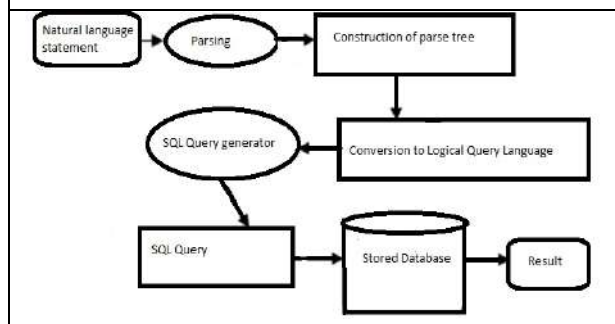
Fig 3. Query in NL to SQL Query conversion
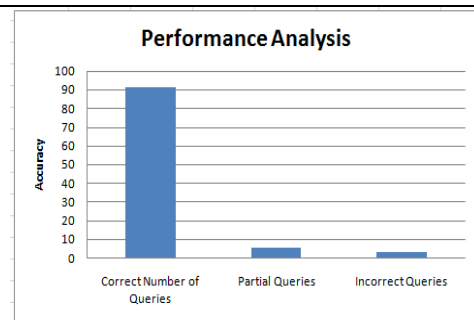
Fig 4. Performance analysis

RESEARCH ARTICLE

Prediction of Cyberspace Shopper's Buying Pattern – a Machine Learning Approach

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ABSTRACT

The ubiquity of the Internet, particularly in developing and underdeveloped countries, is largely responsible for the exponential rise of e-commerce. Many people are taking advantage of online services, such as e-commerce, because the Internet is so easily accessible. In the context of e-commerce, forecasting customer behaviour is becoming increasingly important. By facilitating amore personalised shopping process, it can boost consumer happiness and sales, resulting in higher conversion rates and a competitive edge. This research focuses on a predictive analytics architecture that analyses an online retail dataset using feature engineering and a classification algorithm. The proposed model contains methods for checking the correctness of the study's data points. The study's main purpose is to analyse data and predict the buying intention of the online customer as well as to analyse the machine learning models to predict a pattern. The sequential forward selection approach is used to discover the features that are relevant in classification. The training dataset is fed into various classifiers with the selected variables to evaluate which model is the most suitable and accurate in forecasting buying intention. The machine learning algorithms, such as Generalized linear Regression (GLR), Decision Tree(DT) , Random Forest(RF) and Gradient Boosting Tree(GBT) are used in this study to determine the best model to predict the online buyers behaviour

Keyword: e-commerce, online customer, buying intention, Classifier, feature engineering

INTRODUCTION

E-commerce (Electronic commerce) entails the purchase and sale of goods and services, as well as the transmission of payments and data, over an electronic network, most commonly the Internet. E-commerce is a paradigm shift that has an impact on both marketers and customers, especially in the aftermath of the COVID epidemic. Online shopping has evolved into a new mode of purchase. Online shopping provides a platform on which merchants can publish the goods' commercial attributes, characteristic features, special advantages, and other information, allowing consumers to understand the product's performance, as well as provide feedback on the purchasing process and other information after using the product. Consumers may examine product specifications while browsing online reviews and finding relevant information through online shopping, which has become a significant component of the buying process.

In general, reviews assist consumers in making the best purchasing decisions by allowing them to read online reviews to learn about the quality of the goods, the seller's reputation, service and delivery speed, courier delivery service quality, and other relevant information. Analysing client behaviour is a difficult undertaking, so knowing which method to employ is critical. This paper investigated which factors in online reviews influence consumers' patterns using machine learning algorithms such as Generalized linear Regression (LR), Decision Tree (DT), Random Forest (RF), and Gradient Boosted Tree (GB) are used in this study to determine the best model to predict online buyers' behaviour.

REVIEW OF LITERATURE

E-commerce companies can use this cognitive study to track the usage and attitudes associated with their items and develop appropriate marketing strategies to deliver a personalised shopping experience for their customers, thereby improving their profits[1].The purpose of this study is to investigate the relationship between consumer behaviour and willingness to buy. First, we look for a relationship between changing characteristics such as the environment, organisational factors, individual factors, and interpersonal factors and consumer behaviour to buy things[2]. This work attempts to offer predictive analytics to forecast customer behaviour utilising a behaviour informatics and analytics methodology, to acquire a deeper understanding of consumer behaviour to enable predictive analysis and improve corporate decision making[3]. Researcher reveal a method for identifying potential clients for a retail emporium using machine learning[4] Disruptive technologies like the internet of things, big data analytics, blockchain, and artificial intelligence have transformed how businesses work. Artificial intelligence (AI) is the most recent technological disruptor and has enormous marketing transformation potential. Practitioners all over the world are attempting to determine which AI solutions are most suited to their marketing needs[5].The internet has ushered in the era of online shopping, which has had an impact on ordinary people. As a result, to support the rising expansion of the online market, it is vital to research online buying behaviour and customer satisfaction [6].In this research paper retail sales industry, forecasting future sales is critical for improving corporate operations and increasing profits[9].

DATASET PROPOSED FOR THE STUDY

The following figure 1 shows the proposed dataset which is taken from the UCI Machine Learning Repository. The dataset consists of 12330 instances with 18 attributes. since the proposed dataset contains missing values, it must be pre-processed. The data set consists of selected information from 12330patients. The 18 attributes with one dependent variable and 17 independent variables. The attributes involved are shown in following table1

Heatmap

Features about product-related and information-related duration must be retrieved in order to develop ML models. Choosing the proper and relevant features is an important part of building high-performing machine learning models. ML models perform better at predicting purchase activity for an ongoing session if well-correlated features are chosen. The following Figure 2 shows the correlated heatmap of the above dataset.

Missing value need to ensure that our forecast is valid and accurate, we must execute data imputation to replace the missing values. Missing values may result in incorrect prediction results. One-hot encoding is used for categorical data types. The researcher used the given Imputation methods because the data is absent at random. The missing values in the dataset for the attributes are replaced using a linear regression technique. This will result in an unbiased model parameter estimate. Because the dataset contains missing data at random, Imputation using Mean/ Median value id was applied to all the attributes. The performance of the suggested imputation method is tested using the standard error and Root mean Squared as shown for the Linear Model,Gradient Boosted Trees, Decision Tree, Random Forest are shown the following table 2. The difference between the estimates observation and the true observation is used to calculate standard error.

RESULTS AND DISCUSSION

The analysed the dataset in terms of the hours users mostly visit the website for product related duration and informational duration. The distribution of the number of user interactions on the website by hours is depicted in the following Figure 3. The researcher trained four ML models based on extracted features from the session duration. The ML models are Generalized Linear Regression(GLR), Decision Tree (DT), Random Forests (RF) and Gradient Booster Tree(GBT). The purpose of DT is to learn simple decision rules from data attributes to develop a model that predicts the value of a target variable. RF is a meta estimator that employs averaging to increase predicted accuracy and control over-fitting by fitting several decision tree classifiers on various sub-samples of the dataset. Bagging is an ensemble meta-estimator that trains individual base classifiers on random subsets of the original data set, then aggregates their individual predictions by voting or averaging to generate a final prediction. The GLM extrapolates linear regression by allowing the linear model to be associated to the predictor variables via a logistic function and the magnitude from each measurement's variance to be a factor of its fitted model. GBT is used to limit the risk of over fitting that each individual tree has, integrate multiple decision trees. The researcher conducted experiments for each model to identify which model is performing better and obtained the following results. Prediction Charts
The following figures depict the prediction of product related duration.

As per the comparison table and prediction chart, the researcher identified the best performing model from the above specified Machine Learning model's researcher evaluated for early purchase intention prediction was determined to be the DT classifier. We discovered that product-related duration is the most important feature for purchase prediction based on feature importance analyses. The reason for this could be because product related duration time is a good predictor of users' purchase intent. When a buyer spends longer time on an e-commerce site, for example, it indicates that the customer has a purchase intent, which enhances the likelihood of a purchase action.

CONCLUSION

It is no longer questionable that in the Industrial Revolution, where intelligence will reign supreme, organizations who create excellent customer experiences will prevail. Todays' Industrial Revolution has been envisioned as a corporation with integrated data about customers and products across all channels and products, with that data being used to understand better the end customer experience and visibility across all functional areas. In this context, AI and machine learning have played a critical role in big data analytics, anticipating, and delivering guided experiences that fulfil user expectations. The authors presented a comprehensive picture on applying AI to improve customer experience in this study. The key to providing customer experiences that develop advocacy and customers for life is to use AI and predictive analytics. In this paper, the researcher   evaluating the performance of machine learning models. This could have the added benefit of recommending products and providing discounts to users while they are still logged in. Many studies have shown that offering discounts and product recommendations can help consumers make better purchasing decisions.

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Table -1 Attributes

Attribute No	Attributes	Attribute type	Attribute Code
1	Administrative	Int	Ad
2	Administrative_Duration	Real	Dur
3	Informational	Int	Infm
4	Informational_Duration	Int	inf_Dur
5	ProductRelated	Real	Pro
6	ProductRelated_Duration	Real	pro_Dur
7	BounceRates	Real	Bou
8	ExitRates	Real	Xit
9	PageValues	Int	Pgval
10	SpecialDay	Real	Sp
11	Month	String	Mo
12	OperatingSystems	Int	Os

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| 13	Browser	Int	Bro
14	Region	Int	Reg
15	TrafficType	Int	Tr
16	VisitorType	Real	Vis
17	Weekend	Boolean	Week
18	Revenue	Boolean	Rev

Table 2 . Performance Comparison

Model Category	Root Mean Squared... Number	Standard Deviation Number	Total Time Number	Training Time (1,00... Number	Scoring Time (1,000... Number
Generalized Linear Mo...	0.020	0.001	4960	10.462	2.028
Decision Tree	0.014	0.001	5263	2.595	1.419
Random Forest	0.014	0.001	81873	56.123	114.355
Gradient Boosted Trees	0.012	0.002	38367	35.118	14.193
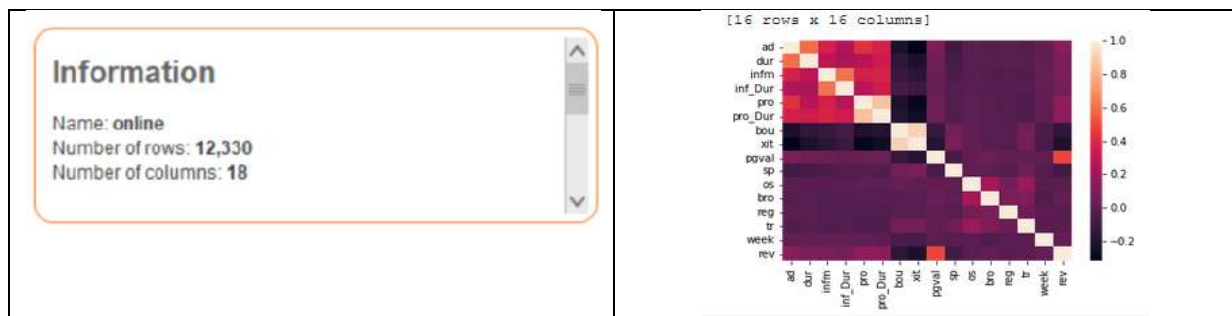
Fig.1 Dataset from UCI Repository

Fig.2 Heatmap
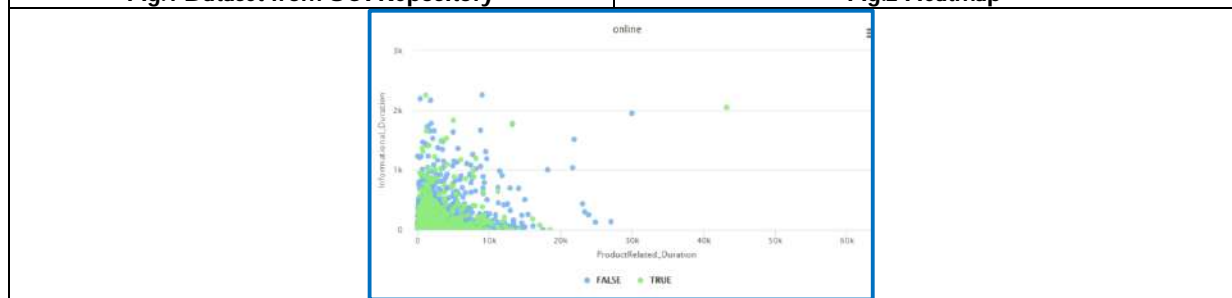
Fig. 3Number of user Interactions
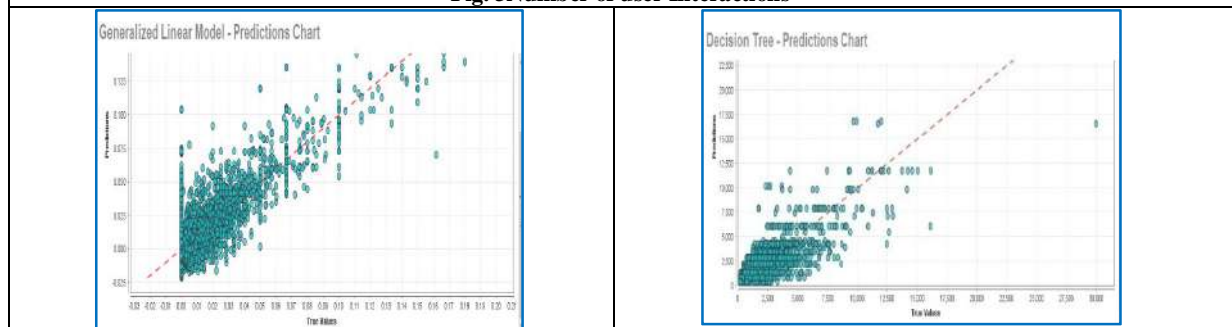
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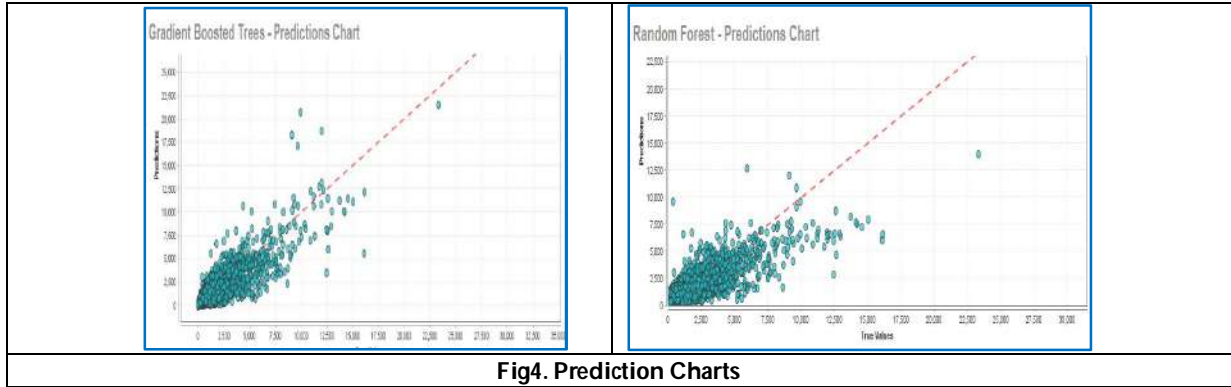
Fig4. Prediction Charts

RESEARCH ARTICLE

Identification of Soil Particles Segmentation using Image Processing

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ABSTRACT

The soil particle has been divergent in every soil texture. The particles have been referred as Sand, Slit and Clay the soil surface has been considered as a most active part of soil particles. The particles in the surface takes and exchange the nutrient content. The nutrient has been essential part to grow crop. Detecting the Sand, Slit and Clay particles from the soil from a very large surface area has been automated in many technological developments. The technology includes IoT has integrate with technology of image processing, datamining etc. to make efficient and smart agricultural development. In this paper the reviews the soil analysis related research to evaluate the efficient improvement over the smart agricultural. Numerous specialists progress checked discernments in the writing to defeat these issues. Without qualified evaluation, utilizing the settled ideas won't meet the normal result of impending analysts. Without a point-by-point audit of momentum indications of progress, the specialists may not discover openings for approaching improvements toward greatness. The understudies turning out to be acceptable specialists require a far-reaching examination of extraordinary ideas with contemporary development. Various works gave in the far-reaching diaries somewhere in the range of 2013 and 2020 are checked on in this paper to achieve these necessities. The checked-on articles are thought about concerning calculations' effortlessness, the sort they have a place, and the exhibition measurements. This survey is relied upon to deliver reasonable reference for getting, choice, and improvements for the understudies, planners, and specialists

Keywords: Technology, IoT, soil, agricultural, crop

INTRODUCTION

Successful agriculture needs a good soil for exchanging and holding nutrient content. The nutrient has been essential part to grow crop. Soil also protects chemical and biological activity especially erosion [1]. The water holding capacity has been determined by Soil particles. The good agriculture practices have been checking the soil analysis before planting soil has been analyzed with its properties involved color, texture and PH value. The particles involved in the Soil has been divided into three group Sand, Silt, and Clay [2]

- **Sand:** The particles of the sand has been measured at 2.0 to 0.05mm.
- **Silt:** The particles of the silt has been smaller than sand 0.05 to 0.002mm.
- **Clay:** The particle has been measured at less than 0.02mm. It has been smallest particles.

Cotton has been considered as an important fiber and increased cost crop in Indian country, it plays a vital role in agricultural industry by increasing economy by its cultivation. Nearly there has been 600 million small farmers in India. Cotton production in India has been considered in world second largest producer in world wide. The productive level has been still below the world average [3][4]. Better management practice has been important for cultivating of various crop that include soil management has been most important part among the other management practices like. Water Management, Test and Disease Management, Harvesting and Storage, Crop Reduce Management, Soil Management etc.[6], Soil Management Practices Include: Soil test based nutrient Identification, Plant nutrient deficiency Identification., Soil Character Identification, Soil Nutrient Identification. One can grow good and produce quality crop by practicing the 4-soil management practice.4 soil management for crop has been considered as the core objective of this work. Successful agriculture needs a good soil for exchanging and holding nutrient content [5]. The nutrient has been essential part to grow all type crop essential for cotton. Soil also protects chemical and biological activity especially erosion. The water holding capacity has been determined by Soil particles. The good agriculture practices have been checking the soil analysis before planting soil has been analyzed with its properties involved color, texture and PH value. The particles involved in the Soil has been divided into three group Sand, Silt, and Clay [7].

Moreover, dry farming is predominant. Therefore, the water and soil conservation measures as well as water harvesting techniques are the need of the hour for getting good crops under dryland farming [8]. Cotton is a major commercial crop of the district and the cotton industry therefore occupies an important place in the economy. Rajapalayam is the important centre for spinning mills and ginning factories. Surgical cotton and bandage cloth are manufactured here. Textile mills in the district produce a variety of cotton yarn [9].

- Development Issues: Popularization of latest water harvesting techniques
Cotton yield must be stepped up further through the application of latest technologies.
- Water and soil conservation measures are to be adopted on large scale.
- Popularization of agricultural machineries especially sowing equipment's
 The smart technology has been increased day-by-day, the smart agricultural has also be essentials in agricultural department. Smart soil analysis also one of the parts in smart agriculture [10].

Soil Test based Nutrient Identification
For cultivating various crop, the nutrient properties have been important. This can be achieved by soil analysis by labor driest. The proposed work has been analysis with various research work done by smart soil analysis by android-based application development by simple soil surface photograph [11][12].

Plant Nutrient Deficiency Identification:
In the natural practice the deficiency has been identified by the cotton cultivation. The proposed work the nutrient deficiency has been identified by the image of the cotton crop [13].

Soil Character Identification
Before cultivating any plant, the character of the soil has been measured for good productivity. It has been done by soil surface image and android application [14].

Soil Nutrient Identification
The nutrient of the NPK level has been identified to enhance the soil by fortifier. The proposal has been considered this has been core work for soil practice management [15].

Image processing and agricultural
Image enhancement is a vast field in digital image processing. There exist many techniques to improve image quality. Histogram equalization is one of the techniques and equalize the grey levels in digital image [16]. This technique is essential for segmentation process. Each object is distinguished from the background by its up-lift edges. Morphological techniques probe an image with small shape or template called structuring element and all the morphological operators are based on this structuring element. These operators are applied directly on watershed segmented image. Watershed transform technique is applied on the gradient image [3]. Three points should be identified for segmentation (i) points belong to regional minimum (ii) catchment basins of regional minimum (iii) watershed lines. The basic steps followed in this method are (a) select original image. (b) Find out the gradient image of image (a). (c) Watershed lines are obtained from image (b). (d) Find out the connected regions (e) watershed lines are obtained from smoothed image (b).

The over segmentation problem in watershed transform is solved by the use of markers. Internal markers are used to obtain watershed lines of gradient image which is to be segmented. The obtained watershed lines are referred as external markers. Therefore, each region has external marker contains internal marker and its background. For this marker-controlled segmentation Sobel operator is used and it's suitable for edge detection. The equation for marker-controlled watershed segmentation is, $M = (\sqrt{M_x2 + M_y2})$. Morphological operations are based on the structuring element. The structuring element is a small shape which positioned at all possible locations in the image and it is compared with the corresponding neighbourhood of pixels [17][18]. Erosion and Dilation are the basic morphological operations which are applied on the segmented brain image. Dilation should be followed by erosion. The erosion operator shrinks the image. The holes and gaps between different regions become larger, and small details are eliminated. The dilation operator grows the eroded image. The holes enclosed by a single region and gaps between different regions become smaller. Results of dilation or erosion are influenced both by the size and shape of a structuring element. Dilation and erosion are *dual* operations in that they have opposite effects. Both dilation and erosion operator denoted by g = f o s. Finally, the area and perimeter of the dilated image are.

In this pre-processing process the input sail surface image has been converted into grey scale image. Segmentation and other process or based on gray scale values. Each pixel contains the value from 0 to 255 various intensity values such as black to white. Resized for convenience. Histogram equalization method has been one of the image enhancement methods used to contrast the image. This is the process of equalizing all the grey levels in the digital image. There are various ways to perform segmentation. Watershed segmentation method has been used to segment the Particle area from input sail surface image [19][20][21]. To overcome the over segmentation problem, the watershed transform method marker-controlled watershed segmentation has been used. It can achieve one-pixel wide, connected, closed and exact location.

Block Diagram
Block Diagram of outline. The basic concept of watershed is based on visualizing a Gray level image into its topographic representation [22]. The two principal morphological operations are dilation and erosion. Dilation allows objects to expand, thus potentially filling in small holes and connecting disjoint objects [23]. Erosion shrinks objects by eroding their boundaries. Dilation usually followed by erosion. The area and perimeter morphological operator are used to find out area and perimeter of the dilated image. Finally, the perimeter is merge into the original input image for highlight the blood clot part clearly [24][25].

Data mining and Agricultural

Information mining is the way toward seeing huge banks of data to create new data. Instinctively, you may imagine that information "mining" alludes to the extraction of new information, yet this isn't the situation; all things being equal, information mining is tied in with extrapolating designs and new information from the information effectively gathered [26]. Depending on procedures and advances from the crossing point of data set administration, insights, and AI, experts in information mining have devoted their vocations to better agreement how to measure and reach determinations from tremendous measures of data. Be that as it may, what are the strategies they use to get this going [27].

Data-Mining Techniques

Information mining is exceptionally powerful, insofar as it draws upon at least one of these procedures:
Possibly the most fundamental procedures in information mining are figuring out how to perceive designs in your informational collections [28]. This is normally an acknowledgment of some deviation in your information occurring at ordinary stretches, or a back-and-forth movement of a specific variable over the long run. For instance, you may see that your deals of a specific item appear to spike not long before the special times of year, or notice that hotter climate drives more individuals to your site [29]. Grouping is a more intricate information mining strategy that constrains you to gather different credits together into discernable classifications, which you would then be able to use to reach further determinations, or serve some capacity. For instance, in case you're assessing information on singular clients' monetary foundations and buy accounts, you could possibly order them as "low," "medium," or "high" credit hazards. You could then utilize these groupings to learn significantly more about those clients [30].

Affiliation is identified with following examples, yet is more explicit to conditionally connected factors. For this situation, you'll search for explicit occasions or characteristics that are profoundly associated with another occasion or trait; for instance, you may see that when your clients purchase a particular thing, they additionally frequently purchase a second, related thing. This is typically what's utilized to populate "individuals likewise purchased" areas of online stores. Much of the time, basically perceiving the overall example can't give you an unmistakable comprehension of your informational collection. You likewise should have the option to distinguish peculiarities, or exceptions in your information. For instance, if your buyers are solely male, however during one peculiar week in July, there's an immense spike in female buyers, you'll need to explore the spike and see what drove it, so you can either recreate it or better comprehend your crowd simultaneously [31].

Bunching is fundamentally the same as characterization, however includes gathering lumps of information dependent on their similitudes. For instance, you may decide to bunch various socioeconomics of your crowd into various bundles dependent on how much extra cash they have, or how regularly they will in general shop at your store [33]. Relapse, utilized essentially as a type of preparation and displaying, is utilized to distinguish the probability of a specific variable, given the presence of different factors. For instance, you could utilize it to project a specific cost, in light of different components like accessibility, shopper interest, and rivalry. All the more explicitly, relapse's primary centre is to assist you with revealing the specific connection between (at least two) factors in a given informational collection [32]. Expectation is quite possibly the most significant information mining methods, since it's utilized to extend the kinds of information you'll find later on [34]. As a rule, simply perceiving and understanding recorded patterns is sufficient to graph a to some degree precise forecast of what will occur later on. For instance, you may audit buyers' records as a consumer and past buys to anticipate whether they'll be a credit danger later on.

Mining Tools

So, do you need the best-in-class AI innovation to have the option to apply these methods? Not really. Indeed, you can most likely achieve some forefront information mining with generally unassuming data set frameworks, and straightforward instruments that practically any organization will have. Also, in the event that you don't have the correct devices for the work, you can generally make your own [47][48].

Review of the Literature

In 2019 [35] a recently planned direct ductile test mechanical assembly was utilized to quantify the elasticity of an unsaturated clayey soil. An advanced picture securing and investigation framework was created for malleable strain examination with the assistance of Particle Image Velocimetry (PIV) and Digital Image Correlation (DIC) methods. Six gatherings of tests were compacted at a dry thickness of 1.7 Mg/m3 and distinctive water substance (6.5%, 8.5%, 10.5%, 12.5%, 16.5% and 20.5%). Test outcomes show that the rigidity trademark bend (elasticity versus water content) of the compacted unsaturated soil displays mono-top element. At the point when water content is generally low, the rigidity increments with expanding water substance and arrives at the greatest incentive at a basic water substance of about 9.3%. At that point, it decreases with additional expansion in water content. The advancement of elasticity with water content relies upon both attractions and microstructure. In light of plotted malleable burden dislodging bends, the pliable disappointment interaction can be separated into three run of the mill stages which are: stress expanding stage (I), disappointment creating stage (II) and post-disappointment stage (III). It is discovered that the generally speaking tractable disappointment measure presents various examples constrained by water content. By and large, the disappointment creating stage (II) endures longer and the disappointment flexibility is more articulated when the example is compacted at higher water content. Utilizing PIV and DIC strategies, the advancement of dislodging course and strain fixation during pressure can be all around caught for enthusiasm for the dirt disappointment system. In view of the strain focus data, the pliable crack area and bearing can be pre-decided for soil tests.

In 2018 [36] investigates the exhibits of utilization on advanced picture preparing for the spatial example acknowledgment and characterisation of Ni focuses in dirt in Europe. Moving normal smoothing was applied to the TIN-introduced framework model to stifle little abnormalities. Computerized picture handling was applied at that point to the matrix. A few NE-SW, E-W and NW-SE arranged highlights were uncovered at the mainland scale. The predominant NE-SW direct highlights follow the Variscan and Alpine orogenies. The most noteworthy changeability zones are in the Alps and the Balkans where mafic and ultramafic rocks outcrop. A solitary significant E-W arranged north-bound component runs along the last mainland glaciation zone. This zone moreover corresponds with a progression of neighborhood maxima in Ni fixation along the glaciofluvial stores. The NWSE prolonged highlights are situated in the Pyrenees, northern Italy, Hellas and Fennoscandia. This investigation shows the upsides of advanced picture handling examination in recognizing and portraying spatial geochemical designs concealed before on regular shading surface guides.

In 2017 [37] The goal of this investigation was to build up an adaptable and free picture preparing and examination arrangement, in light of the Public Domain ImageJ stage, for the division and examination of complex organic plant root frameworks in soil from x-beam tomography 3D pictures. Differentiating root models from wheat, grain and chickpea root frameworks were filled in soil and examined utilizing a high goal miniature tomography framework. A full scale (Root1) was created that dependably related to great to high precision complex root frameworks (10% overestimation for chickpea, 1% underestimation for wheat, 8% underestimation for grain) and gave investigation of root length and point. In-assembled adaptability permitted the client collaboration to (a) change any part of the full scale to represent explicit client inclinations, and (b) take record of computational restrictions of the stage. The stage is free, adaptable and exact in investigating root framework measurements.

In 2020 [38] deals with the vast majority of the past scientists utilized manual picture handling approach through a public space apparatus (Image J) to decipher soil surface dampness content. In any case, the manual handling couldn't be conceivable, when the quantity of pictures is significantly enormous. Also, results couldn't be repeated with regular manual picture preparing. This specialized note acquaints a novel method with computerize the quantification cycle of soil surface dampness content. A stepwise technique was shown to eliminate client reliance for soil shading investigation utilizing a self-ruling Python content. The pictures of the compacted soil were caught utilizing an industrially accessible camera model. The picture examination was directed utilizing customary manual picture preparing approach and recently created procedure. The difference between the mean dim values got from

the previously mentioned two methodologies was low (<3%). Thus, the recently evolved method is cost-effective and plausible for programming with robots to screen soil surface dampness content in huge regions.

In 2018[39] investigated Soil MATTic for quicker and exact soil investigation contrasted and traditional strategy to manage ranchers on crop reasonableness and increment ranch profitability and harvest yield. The Arduino-based model computerized the entire cycle of macronutrient and pH investigation of soil from soil testing techniques up to compost proposal. It incorporates stepper engines and siphons to completely computerize the compound response of soil with synthetic reagent during testing and an on-board printer to print out compost suggestions. It utilizes computerized picture handling strategy to productively distinguish (1) Nitrogen, (2) Phosphorus, (3) Potassium and (4) pH level of Philippine farmlands. The framework is made out of five phases specifically: mechanized soil testing, picture securing, picture handling, preparing framework, furthermore, and suggestion. Counterfeit Neural Network offered quickly furthermore, precise execution for the picture preparing. The framework information base put away and oversees 356 caught pictures where 70% is for preparing, 15% for testing and 15% for approval. Consequences of this examination showed 96.67 precision in distinguishing soil macronutrient and pH level and gives compost proposal for Inbred rice plant, Inbred corn, Tobacco, Sugarcane, Pineapple, Mango, Coconut, Abaca, Coffee, Banana through a produced report in printed structure.

In 2017 [40] deals with the advanced picture handling strategy was utilized to productively recognize the Macronutrients and pH level of Soil in the farmland of Philippines: (1) Nitrogen, (2) Phosphorus, (3) Potassium and (4) PH. The synthesis of the framework is made of four areas to be specific, picture procurement, picture handling, preparing framework, and result. The Artificial neural network was applied in this examination for its highlights that make it appropriate in offering quick and exact execution for the picture preparing. The framework will base on 448 caught picture information, 70% for preparing, 15% for testing and 15% for approval. In light of the outcome, the program will produce a report in printed structure. In general, this examination recognizes the dirt macronutrient and pH level of the dirt and gives manure suggestion for ingrained rice plant and was demonstrated 98.33% precise.

In 2018[41] experimented Digital picture handling has been confirmed to be a function in instrument for investigation in a few fields and applications. In Agriculture area the different boundaries like amount and nature of item are the fundamental measures from the ranchers' perspective. Rural soil is considered as quite possibly the most cherished regular asset whose dirt pH ought to be property used to characterize the degree of causticity or basicity which will influence the supplement accessibility of the dirt and furthermore the development of plants. With the pH Value 7.0, viewed as impartial, the qualities which are above and beneath are either antacid or acidic in nature. This paper includes different techniques what's more, strategies from advanced picture handling to decide and investigate pH esteem from Agricultural soil.

In 2017 [42] deals with a study, the investigation showed that ISS adjusted the efficient changes of the TAU convention to make it more steady; notwithstanding, the insightful outcomes from the TAU convention with ISS didn't show observably higher improvement than the CULS (non-ISS) yields. It appears to be that keeping a convention (regardless of what the convention is) consistent, decreases the productivity of ISS, as the non-efficient impacts are negligible. In spite of the fact that, utilizing ISS is energetically suggested for adjusting little deliberate impacts in a given convention as a rule and between conventions specifically. As far as information mining methods, PARACUDA II® as another information mining and model improving methodology, clearly gave better outcomes and end up being an incredible and solid instrument in accomplishing the best expectation model, instead of PLSR as a customary information mining calculation; notwithstanding, PARACUDA II® is additionally a PLSR model, yet it utilizes a covariate enhancement schedule that chooses the best pre-processing ventures for a given reaction variable. In view of the promising consequences of the PARACUDA II® execution, usage of additional examinations with other information mining and AI calculations over various geographic scales is exceptionally upheld to check the PARACUDA II® strength and dependability. All in all, adherence to a steady convention and a solid information mining strategy will improve the exactness and the likeness of results, which is fundamental for the whole soil local area.

In 2018[43] This work has the reason for building up a system utilizing multi-transient satellite picture data, meaning to develop a solitary manufactured picture which would address soils. The work was completed in the territory of São Paulo, Brazil, on a site covering 14,614 km2 The strategy, assigned as Geospatial Soil Sensing System (GEOS3), depends on the accompanying advances: a) formation of an information base with Landsat 5 inheritance information.; b) sifting of the data set to give pictures just from the dry season in the locale; c) inclusion of a bunch of rules into the framework to channel different articles other than soils; d) Each uncovered soil event for every area along the time-arrangement was utilized to figure a Temporal Synthetic Spectral Reflectance (TESS) of the dirt surface; e) accumulation of all TESS creates the Synthetic Soil Image (SYSI); f) quantitative and subjective approval of the SYSI through the relationship among' s lab and TESS, soil line evaluation and the main segment examination (PCA). GEOS3 had the option to give the best delegate reflectance of soils for each band during the chronicled period

Accordingly, TESS isn't the 'valid' yet a manufactured unearthly reflectance. The sanctioned relationship among' s research centre and satellite information arrived at 0.93. An estimation of up to 0.88 in the Pearson's connection among' s research facility and TESS was additionally accomplished. In a solitary scene, just 0.5% of region was accessible as separated uncovered soil for spatial examination. Notwithstanding, SYSI arrived at 68%. Considering just the sugarcane horticultural zones, an estimation of 92% was accomplished. Our examination shows that a multi-worldly information mining strategy can recover soil surface portrayal. The way in to the outcomes was figuring the middle otherworldly reflectance from the uncovered soil pixels along the time of the time arrangement. GEOS3 items can help soil assessment by aiding computerized soil planning, soil security, exactness farming, soil quality measurement, soil preservation, and climate observing and soil test distribution, among others

In 2017 [44] Farming is the main application region especially in the non-industrial nations like India. Utilization of data innovation in farming can change the circumstance of choice making and ranchers can yield in better manner. Information mining plays a pivotal job for dynamic on a few issues identified with horticulture field. It examines about the part of information mining in the farming field and their connected work by a few creators in setting to farming area. It likewise examines on various information mining applications in addressing the distinctive farming issues. In this study it came to realize that the k-implies calculation is utilized for soil characterizations utilizing GPS-based advancements, Classification of plant, soil, and build-up locales of interest by shading pictures, Detecting weeds in exactness farming.

In 2017 [45] This investigation has inspected 66 long haul exploratory correlations on Soil Organic Carbon (SOC) and tillage frameworks in Mediterranean arable harvests (from 15 destinations situated in Greece, Italy, Morocco and Spain), with the intend to recognize the biophysical and agronomic factors most connected with C sequestration rate. Information was coordinated in a dataset containing essential ecological descriptors (rise, temperature, precipitation), data on soil culturing framework (ordinary, least, no-culturing), soil credits (pH, molecule size conveyance and surface), crop revolution, preparation, time length of the investigation, starting and last SOC stocks. The gathered data were dissected utilizing an information mining approach including Spearman non-parametric relationships, Principal Component Analysis (PCA), progressive grouping and step-wise numerous relapses. Culturing, crop revolution, what's more, treatment were the main variables influencing C sequestration rate. Non-parametric relationships revealed negative coefficients for starting SOC stock, length of the investigation, mineral preparation, culturing and creation framework. C sequestration rate expanded altogether under no-culturing. Various levelled bunching demonstrates that geological closeness reflects similitude in biophysical conditions and agronomic rehearses. PCA illustrated a positive relationship of SOC with soil profundity, height and locales situated in Spain and a negative connection with mean air temperature, mineral treatment, water system, examination's length and locales Situated in Greece. C sequestration rate was decidedly related with mean air temperature. At long last, a stepwise numerous relapses showed that C sequestration rate expanded in locales presented to colder environment conditions and under no-culturing

In 2020 [46] deals with the investigation enhances past computerized planning of soil thickness across Australia utilizing a methodology fit to the landmass' one of a kind pedo-geomorphic history. Utilizing three enormous, in situ perception datasets and a wide scope of spatial ecological factors, we created three models portraying rock outcrops, middle of the road and profound soils individually. Our demonstrating approach tended to right-edited information, which is a typical quality of soil thickness information, and we applied an iterative, information re-examining system to evaluate forecast vulnerabilities. This paper incorporated the three models to make soil thickness maps and related results of soil thickness exceedance probabilities. Utilizing information rejected from model alignments, we accomplished a general exactness of 99% for the parallel result rock outcrops model, and 85% for the double result profound soils model. Demonstrating soil thickness of shallow to profound soils brought about a concordance coefficient of 0.77. Of the relative multitude of ecological factors considered in this examination, those related with environment information (counting topo-environment) were reliably the most regularly utilized and significant. We partner this finding with the immediate and circuitous impacts of environment on biota and enduring of parental materials alongside different components driving spatial heterogeneity in soil thickness across Australia. While the items produced by this exploration are not without mistake, the general example of soil thickness is predictable with past perceptions from authentic soil overviews across Australia and the outcomes are evidently more skilful than past advanced soil planning endeavours.

CONCLUSION

Soil particles detection from soil surface image has been developed by using image processing and datamining techniques has been evaluated Both techniques have been very useful to extract the various particles present in images. Future to develop a comparison of other segmentation methods and various morphological operators. The segmented particles have been classified by using classification algorithm with real time soil analysis

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Table 1. Particle type, number of particles per gram, and the average surface area per gram.

[1] **Particle Type**	[2] **Diameter (mm)**	[3] **Number of Particles per Gram**	[4] **Specific surface area (cm /g)**
[5] **Clay**	[6] < 0.002	[7] 90,260,853,000	[8] 8,000,000
[9] **Rough sand**	[10] 1.00-0.50	[11] 720	[12] 23
[13] **Well sand**	[14] 0.25-0.10	[15] 46,000	[16] 91
[17] **Average sand**	[18] 0.50-0.25	[19] 5,700	[20] 45
[21] **Silt**	[22] 0.05-0.002	[23] 5,776,000	[24] 454
[25] **Very rough Sand**	[26] 2.00-1.00	[27] 90	[28] 11
[29] **Very well sand**	[30] 0.10-0.05	[31] 722,000	[32] 227

Fig. 1. Block Diagram

RESEARCH ARTICLE
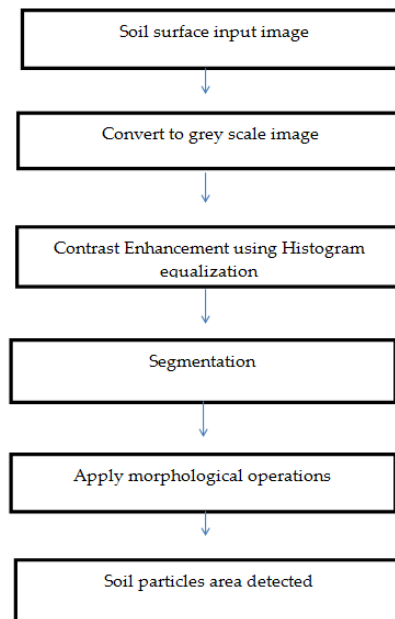
Cascade Classifier and Local Binary Patterns Histograms (LBPH) Based Face Authentication Smart Attendance System (FASAS)

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ABSTRACT

Even though they are sophisticated and time-consuming for monitoring regular attendance logs, attendance management systems are very crucial to all organizations. Face identification has been a popular study topic in recent years since it is used as a biometric verification method in a variety of applications, including attendance management and access control systems. Face Authentication Smart Attendance System (FASAS) is introduced in this study, which is based on biometric-driven facial scans. The FASAS system intends to provide a class attendance system based on the face authentication idea. The four processes of the FASAS system are database creation, face detection, face authentication, and attendance updating. Images of pupils in class are used to construct a database, which is organized by name and registration number. Face detection is carried by using the Haar Cascade technique. A feature tensor matrix is created from the retrieved features. Using the Local Binary Patterns Histograms, the feature matrix of the key face picture is compared to the feature matrices of the target face image that are recorded in the feature vector database for face authentication (LBPH). Faces are recognized and authenticated from the classroom's live streaming footage. At the end of the session, the student's attendance will be generated as a Microsoft Excel (MS) sheet and emailed to the appropriate faculty. The proposed method produces results that are comparable to those achieved by existing methods.

INTRODUCTION

Face recognition is one of the most well-known biometric technologies. Human–computer interaction, face tracking, facial expression recognition, fatigue detection, pedestrian and upper body detection [1,2], visual surveillance, gesture recognition [3, robotics, video and image indexing, driving monitoring system, and multimedia and forensic applications are just a few of the application domains where it is used. Face recognition has emerged as a feasible biometric recognition approach in the current context of heightened security. Face recognition methods are divided into two categories: geometrical-driven or model-based methods [4,5] and appearance-based or statistical method-based methods [6,7]. Face shape cues such as the width of the mouth, inner and outer distances between two eyes, and heights between the forehead and nose and chin were used to build geometrical-driven techniques. Low-level data such as colour, texture, and spatial orientation have been used to construct appearance-based techniques [8,7]. For this reason, numerous existing solutions exist, such as biometric attendance and RFID using an Attendance system. All of these approaches are time-consuming since students must form a line to place their thumb on the scanning gadget. Those gadgets are extremely pricey.

The purpose of this study is to design an attendance system that uses face verification to automatically indicate student attendance. Taking students' attendance in the classroom is a crucial duty that, when done manually, takes a long time when the number of students is large. This project will recognize students' faces and keep track of their attendance [14,15]. The Haar-cascade classifier is used to categories and obtain the true face of a student. It crops the student's face while removing the undesired background, which is important in face classification. Local Binary Patterns Histograms (LBPH) is a face recognition algorithm that checks if the user's face exists in the dataset and displays the student's name and enrollment number if it does. Facial Authentication Smart Attendance System (FASAS) is a project that aims to create a more cost-effective and efficient automated student attendance system using face recognition. The outcome will be displayed in a Microsoft Excel spreadsheet.

LITERATURE REVIEW

Muqeet and Holambe [8] proposed a new face feature extraction approach based on DIWT and LBP. They've also devised a quadtree partitioning mechanism, which makes adaptive direction selection in local regions for DIWT sub-bands easier. To produce a local descriptive feature set, LBP histogram features are extracted from selected top-level DIWT sub-bands**.** Sharifara *et al.* [9] use the extended local binary pattern (LBP) and support vector machines with Adaboost algorithm to discover Haar-like texture features and then validate the method. Non-facial objects that may appear during the face detection process are rejected using a validation process. Extended Local Binary Patterns are used in a two-stage validation process (ELBP).

Geetika and Indu [10] provide a framework that extracts Orthogonal Combination of LBP (OC-LBP) and Histogram of Oriented Gradients (HOG) features, normalises them, and then fuses them together. Following that, classification is accomplished using a histogram-based Chi-square, Square-chord and extended-Canberra metrics, as well as SVM using a normalised Chi-square kernel. Moeini *et al.* [11] proposed a method for recognising look-alike faces in which people's facial depth is not variable and inter-class similarity is greater than intra-class variation. To solve this difficulty, they use three alternative ways to extract deep and textural characteristics, including sparse, collaborative representation, and a hybrid of the two. Then, for each individual in the gallery, each 3D reconstructed face is synthesised into many alternative viewpoints, and a sparse dictionary is developed based on the synthesised face picture.

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Zangeneh and Moradi [12] described by the technique is based on the differential geometric feature points of the nose, eyebrows, and mouth. To recognise the face photos, they used an SVM classifier. The distance between the important points of the face and the reference point was determined in both x and y directions for two photographs in this method, and the difference between the distances was used to derive the differential geometric features between the two images. Vasanthi and Seetharaman [13] introduced a multivariate correlation analysis-based technique for biometric-driven facial image recognition that extracts geometrical feature points and low-level visual information. Low-level visual elements like colour and texture are extracted at the local level from a few significant sections of the facial image. The Active Shape Model is used to capture the geometrical aspects (ASM). The colour characteristics are taken from the YCbCr colour model, while the texture features are extracted using the autocorrelation approach. A feature tensor matrix is created from the retrieved features. Using the Canonical Correlation approach, the feature matrix of the key face picture is compared to the feature matrices of the target face image stored in the feature vector database. It is determined whether the association between the key and target feature matrices is highly significant or not. If the correlation is significant, it is assumed that the key and target face photos are the same; otherwise, it is assumed that they are not.

Proposed System

Face Authentication Smart Attendance System (FASAS) is introduced in this study, which is based on biometric-driven facial scans. The FASAS system intends to provide a class attendance system based on the face authentication idea. The four processes of the FASAS system are database creation, face detection, face authentication, and attendance updating. Images of pupils in class are used to construct a database, which is organised by name and registration number. Face detection is carried by using the Haar Cascade technique. A feature tensor matrix is created from the retrieved features. Using the Local Binary Patterns Histograms, the feature matrix of the key face picture is compared to the feature matrices of the target face image that are recorded in the feature vector database for face authentication (LBPH). Faces are recognised and authenticated from the classroom's live streaming footage. At the end of the session, the student's attendance will be generated as a Microsoft Excel (MS) sheet and emailed to the appropriate faculty.

Dataset Creation

A web cam is used to collect images of students. Multiple photographs of a single student will be taken from various angles and gestures. Pre-processing is applied to these photos. Cropping the photos yields the Region of Interest (ROI), which will be used in the recognition procedure. The cropped photos are then resized to a specific pixel point. Following that, these photos will be transformed from RGB to grayscale. The photographs will subsequently be saved in a folder with the names of the students.

Face Detection

ace detection is done with Open CV and the Haar-Cascade Classifier. Before it can be utilised for face identification, the Haar Cascade algorithm must be taught to recognise human faces [14,15]. Feature extraction is the term for this process. The xml file haar cascade frontal face default was used to train the haar cascade. It is carried out in the following manner:

Take Images: This keeps track of a student's information. This is the project's first phase. Students fill out this form with their name and registration number. It will take student photographs and save them in the Training images folder.

Train Images: This is used to categorise the image according to its intensity. All of this was accomplished with the help of the haar-cascade feature. It removes the unneeded backdrop from the photograph and crops the face from the usual image.

Haar cascade classifiers: Using Haar feature-based cascade classifiers to recognise objects is an effective strategy. It's a machine-learning approach in which a cascade function is learned using a large number of positive and negative photos. After then, it's utilised to find items in other photos. To train the classifier, the method requires a large number of positive images (images of faces) and negative images (images without faces). Then there's the matter of

extracting features from it. Haar characteristics, as illustrated in the graphic below, are employed for this. They're similar to convolutional kernels. Each feature is a single value calculated by subtracting the sum of pixels beneath the white rectangle from the total of pixels beneath the black rectangle (See Figure 1). To calculate a large number of features, all feasible sizes and positions of each kernel are now used. The total of the pixels under the white and black rectangles must be found for each feature computation. They used the integral image to solve the problem. It simplifies the calculations for a specific pixel to a four-pixel process, no matter how vast your image is. Nice. It speeds up the process. However, the majority of the traits we calculated are irrelevant. Take a look at the illustration below. Two good qualities may be seen in the top row. The first attribute chosen appears to be the fact that the area around the eyes is frequently darker than the area around the nose and cheekbones. The second feature chosen is based on the fact that the eyes are darker than the nasal bridge. However, the application of the same windows to the cheeks or any other location is irrelevant. Adaboost is the one who accomplishes it. In table 2, the Haar Features for facial images are explored.

To do so, apply each feature to all of the training photographs. It determines the appropriate threshold for each feature to identify the faces as positive or negative. There will undoubtedly be errors or misclassifications. Choose the characteristics with the lowest error rate, which indicates they classify the face and non-facial photos the most accurately. (The procedure is not as straightforward as this.) At first, each image is assigned the same weight. The weights of misclassified photos are increased after each categorization. After that, the same procedure is followed. Error rates are calculated at new levels. There are also new weights. The method is repeated until the requisite accuracy or error rate is met, or until the required number of features is discovered. A weighted sum of these weak classifiers yields the final classifier. It's named weak since it can't classify the image on its alone, but when combined with others, it creates a strong classifier. round 6000 characteristics were included in their final arrangement. Take a look at each 24x24 window. Apply 6000 characteristics to it. The non-face region of an image makes up the majority of the image. As a result, having a simple method to check if a window is not a face region is a better approach. If it isn't, toss it out in one go and don't bother processing it again. Instead, concentrate on areas where a face might appear. Spend more time checking possible face regions this way. They came up with the idea of a Cascade of Classifiers to do this. Rather of applying all 6000 features to a single window, they are divided into stages of classifiers and applied one by one. If a window fails the first test, it should be discarded..

Don't think about the remaining characteristics. Apply the second stage of features and continue the procedure if it passes. A face region is a window that passes through all stages. The detector has around 6000 features and 38 stages, with the first five stages consisting of 1, 10, 25, 25, and 50 features. According to the authors, each sub-window is examined on average 10 attributes out of 6000+.

Detect: This was once used to take class attendance. The student must first enter their hour details, following which, if the student's face is recognised, the student's name and number, as well as the date and time, will be saved. The information is kept in Microsoft XLsheets.

Performing The Face Authentication

The algorithm has already been trained at this point. Each image from the training dataset is represented by a different histogram. So, given an input image, repeat the processes for this new image, and a histogram representing the image is created. To identify the image that matches the input image, simply compare the histograms of the two images and return the image with the closest histogram.

The Local Binary Pattern (LBP) is a basic yet effective texture operator that labels pixels in an image by thresholding the pixels' immediate surroundings and treating the output as a binary number. It was first described in 1994 (LBP) and has since been discovered to be a useful texture categorization trait. On some datasets, it has also been discovered that combining LBP with the Histograms of Oriented Gradients (HOG) descriptor boosts detection performance significantly. Face images can be represented using a simple data vector using LBP and histograms.

Because LBP is a visual descriptor, it may also be utilised for face recognition tasks, as seen in the steps below. There are four parameters in the LBPH:

• **Radius:** the radius represents the radius surrounding the centre pixel and is used to generate the circular local binary pattern. Normally, it is set to 1.

• **Neighbors:** the total number of sample points used to construct the circular local binary pattern. Keep in mind that the higher the computational cost, the more sample points you include. It's commonly set to eight.

• The number of cells in the horizontal direction (Grid X). The larger the dimensionality of the generated feature vector, the more cells there are in the grid and the finer the grid is. Normally, it is set to 8.

• Grid Y (vertical): the number of cells in the grid. The larger the dimensionality of the generated feature vector, the more cells there are in the grid and the finer the grid is. Normally, it is set to 8.

Training the Algorithm: First, the algorithm must be trained. To do so, you'll need a dataset containing the faces of the persons you wish to recognise. Each image must also include an ID (which might be a number or a person's name) so that the algorithm can recognise an input image and provide you with an output. The same ID must be used for all images of the same individual. Let's look at the LBPH computational steps now that the training set has been built.

Applying the LB Poperation: Here, LBPH is used to create an intermediate image that better describes the original image by emphasising the face features. The algorithm accomplishes this by employing a sliding window notion based on the parameters, radius, and neighbours. This approach is depicted in Figure 3.

o Let's break it down into little steps based on the graphic above to make it easier to understand:

o Assume you have an in grayscale image of a face.

o Create a 3x3-pixel window out of a portion of this image.

o It can also be represented as a 3x3 matrix containing the intensity of each pixel(0~255).

o The central value of the matrix must then be utilised as the threshold.

o The new values from the 8 neighbours will be defined using this value.

o Set a new binary value for each neighbour of the central value (threshold). Set 1 for values that are equal to or higher than the threshold, and 0 for those that are below.

o The matrix will now only include binary values (ignoring the central value). Each binary value from each point in the matrix must be concatenated line by line into a new binary value (e.g. 10001101).

o Next, transform this binary value to a decimal number and assign it to the matrix's central value, which is a pixel from the original image.

o Have a new image at the end of this method (LBP procedure) that better represents the original image's qualities.

Bilinear interpolation can be used to accomplish this. If a data point falls between the pixels, the value of the four closest pixels (2x2) is used to estimate the new data point's value (See Figure 4).

Extracting the Histograms: Now, using the image created in the previous step, divide the image into various grids using the Grid X and Grid Y parameters, as shown in Figure 5 Each histogram (from each grid) will only have 256 places (0255) reflecting the occurrences of each pixel intensity because the image is grayscale. Then you'll need to concatenate each histogram to make a new, larger one. In the final histogram, assuming we have 8x8 grids, this is translated to 8x8x256=16.384 places. The features of the original image are represented by the final histogram.

Attendance Updation: The recognised faces will be marked as present on the excel sheet, while the others will be marked as absent, and the list of absentees will be mailed to the relevant faculties. At the conclusion of each month, faculties will be updated with a monthly attendance sheet.

Implementation

This attendance system is a user-friendly, graphical user interface (GUI) based application that automates attendance activity. The Quality Assurance system includes options for entering data by interacting with user-friendly input

windows, as well as reports. The system will assist in saving time and effort when it comes to preparing attendance and creating essential reports. It ensures that reports are generated without errors.

System Implementation

The system implementation stage of a project is when the theoretical design is translated into a practical system. The final installation of the package in its real environment, as well as the satisfaction of the intended users and the system's operation, is referred to as software implementation. In many organisations, the software development project will be commissioned by someone who will not be operating it. People are unsure whether or not the software is intended to make their jobs easier. They have reservations about the programme at first, but they must guarantee that resistance does not build up, as this must be avoided.

• The active user must be aware of the system's advantages.
• Their trust in the programme must be built up.
• The user is given clear instructions so that he can utilise the application with ease.
• Before seeing the system, the user should be aware that the server software must be operating on the server in order to view the results. The real processes will not take place if the server object is not up and running on the server.

Input Design: The link between the information system and the user is the input design. It entails creating data preparation specifications and procedures, as well as the steps required to convert transaction data into a usable format for processing. This can be accomplished by inspecting the computer to read data from a written or printed document, or by having people key the data directly into the system. Limiting the amount of input required, controlling errors, avoiding delays, avoiding superfluous stages, and making the process simple are all goals of input design. The input is created in such a way that it gives security and convenience while maintaining privacy. There are various forms of inputs in these applications.

For example, there are two sorts of inputs on the student enrollment page: enrollment number and student name. Enrollment No and Student Name are text input fields that will take the student's registration number and name (See Figure 6). Figure 7 shows the training and testing images utilised for the attendance system after the dataset was collected.

Output Design: A high-quality output is one that satisfies the end-user's needs and delivers information clearly. Any system's processing results are conveyed to users and other systems via outputs. It is decided how the information will be displaced for immediate use, as well as the hard copy output, in output design. It is the user's most essential and direct source of information. The system's relationship with the user is improved via efficient and intelligent output design. The output design specifies the output that must be produced as well as the format in which it must be produced. It's crucial to convey the correct information so that the proper decisions can be made. Screen output, output to be stored as files on storage media, and hard copy of the output are the three basic forms of output generated. The screen output is essentially a screen display of the generated output. Most query results are normally presented on the screen to provide on-line information. The provision of created output for storage in a file is a further reference, and the taking of hardcopies of the same is to send information to management and whatever circumstance requires it. The output design objectives are to design output to serve the specified purpose, design output to fit the user, supply the proper quantity of output, and ensure that output is delivered where it is required. Figure 9 shows the attendance result for each student as an excel sheet.

CONCLUSION AND FUTURE WORK

Authentication by Face The Smart Attendance System (FASAS) is designed to provide an easy and convenient environment for automatically taking attendance. It will make the attendance work easy for the personnel. Students who are registered in this system will have their attendance taken automatically by the system. The

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outcome will be displayed in a Microsoft Excel spreadsheet. In this method, enrolling students is likewise a simple process. This technique only requires students to provide their roll number and name in order to take a snapshot. This can be used in a variety of situations, such as days when attendance is tough to come by compared to other days. This project improves the accuracy of attendance work. I intend to take my idea to a larger scale, similar to that of an entire institution. To check a large number of user details and faces at the same time, a more efficient classifier and algorithm are used to create a database to record student details. This type of device protects the school against illegal visitors. It aids in the identification of those individuals. Face recognition technology like this is already in use by the Chinese government in public sectors. These advanced levels are beneficial in a variety of situations, such as recognizing criminals in public settings.

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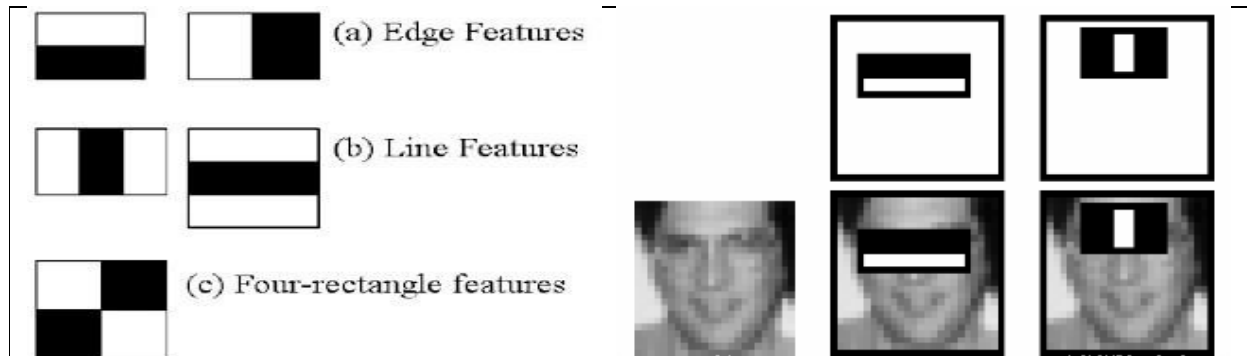
Figure 1. Haar cascade classifiers for features

Figure 2. Haar cascade classifiers for face images
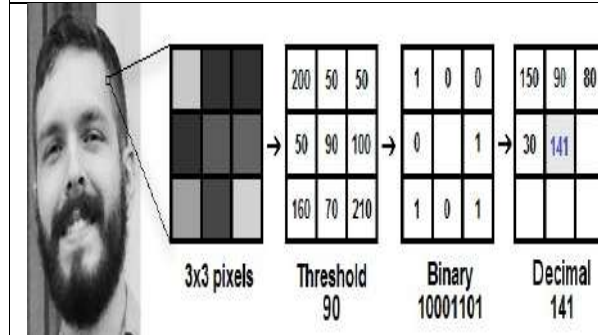
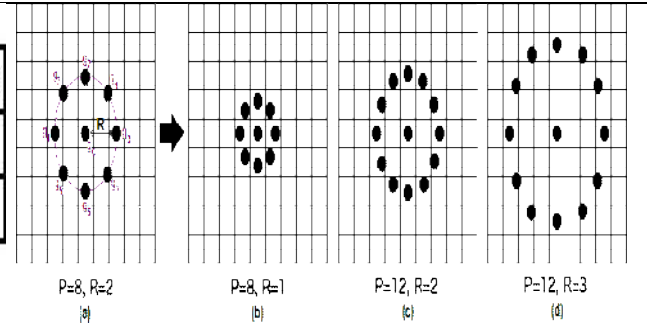**Figure 3. LBPH algorithm for face images**

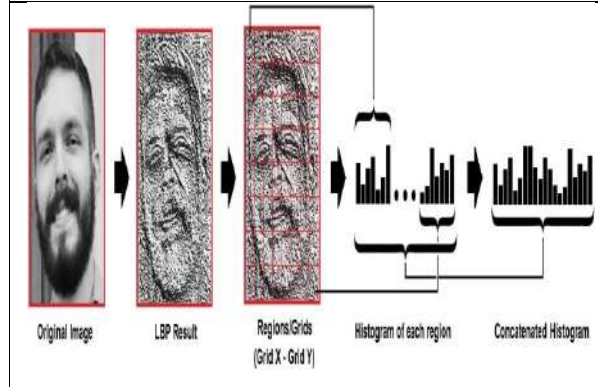**Figure 4. Bilinear interpolation algorithm for face images**
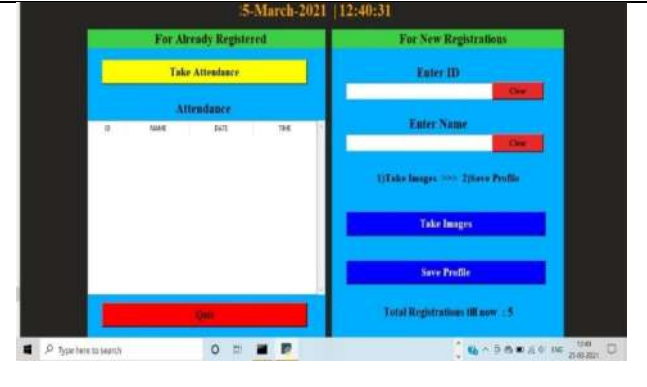

Figure 5. Extracting histogram for face images
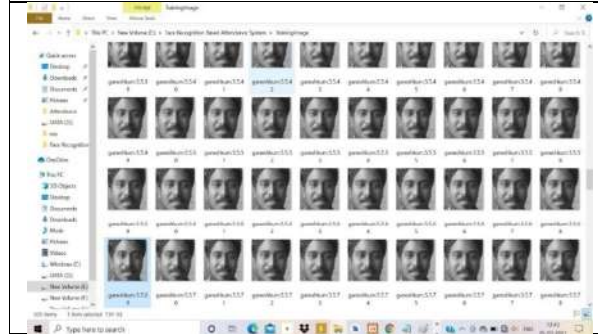

Figure 6. Input design for student entry

Figure 7. Taking & training images for student entry

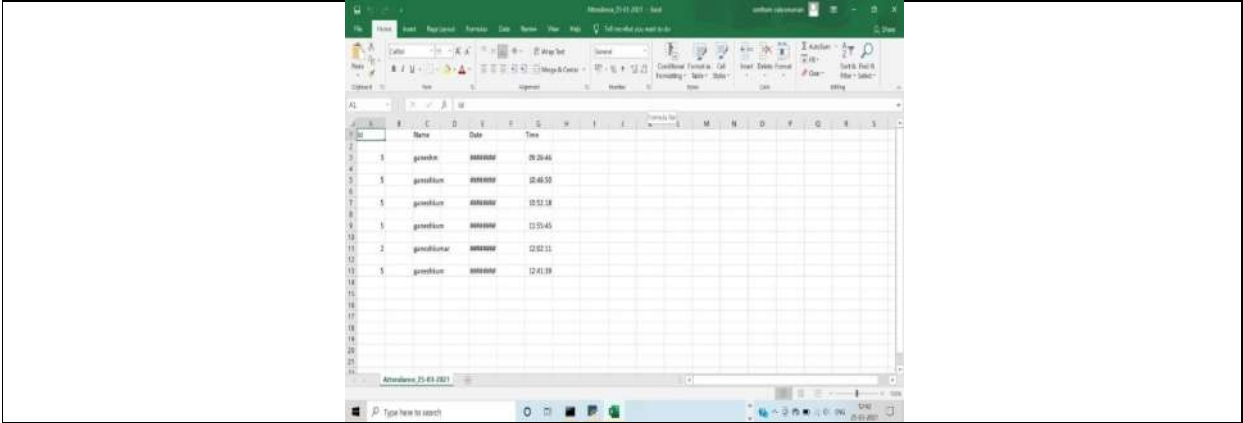**Figure 8. Taking attendance via webcam**

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Figure 9. Output design of excel sheet

RESEARCH ARTICLE

Synergistic Antibacterial Activity of the Probiotic Kefir with Conventional Antibiotics

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ABSTRACT

Kefir is a fermented milk drink prepared by inoculating cow, goat or sheep milk with kefir grains. Kefiris highly nutritious in containing various amounts of dietary minerals, vitamins and essential amino acids. The kefir grain microorganisms produce lactic acid, antibiotic and bactericides, which inhibit the growth of pathogenic microorganisms in kefir milk. Therefore Kefir is considered to be probiotic both for its nutritional value and antimicrobial mechanism. The purpose of this study was to determine the antibacterial activity of Kefir milk in synergy with antibiotics and to fractionate the bioactive compounds present in it.

Keywords: Kefir, probiotic, kefir milk, Kefir grains.

INTRODUCTION

A probiotic may be a single strain or a mixture of different organisms and is believed to improve the wellbeing via the pathological process involving immunomodulatory, metabolic and barrier activities. Kefir or bulagors (in Latin America), is a fermented milk drink similar to a thin yoghurt that is made with a yeast and bacterial fermentation starter of kefir grains. It was originated from Eastern Europe and Russia, where it is prepared by inoculating cow, goat or sheep milk with kefir grains. Kefir is nutritious and considered probiotic [1]. Kefir grains are a kind of yoghurt starter, which are white to yellow-white, gelatinous, and variable in size. Fermentation of lactose yields a sour carbonated, slightly alcoholic beverage with a consistency and taste similar to thin yoghurt. The kefir grains initiating the fermentation are combination of lactic acid bacteria and yeast in a protein, lipid and sugar matrix.

Kefiran a water soluble polysaccharide, imparts a creamy texture to kefir. The grains range in the colour from white (colour of healthy grain) to yellow [2]. Kefir contains various amounts of nutrients including dietary minerals, vitamins and essential amino acids. Minerals found in kefirare calcium, iron, magnesium, potassium, sodium, copper, manganese and zinc. Essential amino acids include methionone, cysteine, tryptophan, phenylalanine, tyrosine, leucine, threonine, lysine and valine. Vitamins include Vitamin A, B_1, B_2, B_3, B_6, B_{12}, C and E[3]. Kefir grains have a diverse microbial species composition such as the predominance of lactic acid bacteria, acetic acid bacteria, yeast and fungi. However depending upon the kefir origin, the substrate used in the fermentation process and the culture maintenance method, the microbial population in kefir may differ. Kefir is primarily considered a probioticaid due to its composition. Probiotics are microbial cell preparations or components of microbial cells with a beneficial effect on the health of the host. Kefir grain microorganisms produce lactic acid, antibiotic and bactericides, which inhibits the formation of pathogenic microorganisms in kefir milk [4].SarikaLokhande*et al*[5]in the year 2018 reported that hydrogen peroxide produced by Lactobacillus, when transferred to an anaerobic environment can also inhibit pathogenic microorganisms.

Several studies proved kefir and its constituents to have improved lactose digestion in addition to their antimicrobial, antitumor, anticarcinogenic and immunomodulatory activity [6]. It also has the potential to heal wounds [7]. Schneedorf JM et al.[8] in 2004 had showed that generally kefir grains contain a relatively stable and specific microbiota enclosed in a matrix of polysaccharides and proteins and that microbial interaction in kefir are complex due to the composition of kefir grains. *In vitro* experimental studies performed so far to understand the antimicrobial mechanism of kefir's microbial flora are limited and therefore this present study is significant. This study examined the antimicrobial activities of kefir using agar diffusion experiments and also fractionating the bioactive components which were responsible for the antimicrobial activity.

AIM AND OBJECTIVES

AIM
To determine the antimicrobial activity of kefir and to fractionate the bioactive compounds present in it.

OBJECTIVES
- Production of kefir.
- Testing the antibacterial activity of kefir using kefir pellet and supernatant.
- To determine the synergistic effect of kefir with antibiotics.
- To fractionate the bioactive compounds in kefir.

MATERIALS AND METHODS

- Kefir grains were bought from the online market Natures Probiotics.
- Bacterial cultures – *Staphylococcus aureus, Escherichia coli, Bacillus cereus, Pseudomonas* sp, *Klebsiella*sp, *Proteus* sp,.
- Muller Hinton Agar for antimicrobial activity.
- Thin Layer Chromatography.

Phase 1: Production of Kefir from Kefir Grains
The milk was poured into a clean glass jar (not metal) and a wooden spoon was used to stir. (The milk could be cold or at room temperature). The jar was covered with cheese cloth and a rubber band was tied to it. (container should not be tightly closed as it would the build-uppres sure during fermentation causing the jar to burst). The container was kept away from direct sunlight at room temperature (ideally around 70°F) away from direct sunlight. The jar was checked every few hours. Kefir was ready for use when the milk got tightened and tasted tangy. (This will usually take about 24 hours at average room temperatures; the milk will ferment faster at warmer temperatures and

slower at cool temperatures. If after 48 hours, the milk has not fermented, the grains are strained out and checked again in a fresh batch, this often occurs when using new kefir grains or when refreshing dried kefir grains, or when using refrigerated grains.

Phase 2: Treating bacterial culture with kefir

➤ Pure bacterial cultures (*Staphylococcus aureus, Escherichia coli, Klebsiella sp., Pseudomonas sp., Bacillus subtilis., Proteus sp.,*) were isolated from their respective selective medium and sub cultured in a sterile Nutrient broth and incubated at 37℃ for 6 hours.

➤ Muller Hinton agar was prepared and sterilized and poured on to the sterile petri plates.

➤ After 6 hours of incubation, the bacterial cultures were lawn cultured onto the solidified Muller Hinton agar plates with a sterile swab.

➤ The wells were cut in the agar using a well cutter aseptically.

➤ Kefir(150 µl)was poured into the wells using a sterile pipette.

➤ The plates were incubated at 37℃ for 24 hours.

Phase 3: Treating kefir'ssupernatent and pellet separately against bacterial cultures

➤ Pure bacterial cultures (*Staphylococcus aureus, Escherichia coli, Klebsiella sp., Pseudomonas sp., Bacillus subtilis., Proteus sp.,*) were isolated from their respective selective medium and sub cultured in a sterile Nutrient Broth and incubated at 37℃ for 6 hours.

➤ Muller Hinton agar was prepared and sterilized and poured on to the sterile petri plates.

➤ After 6 hours of incubation, the bacterial cultures was lawn cultured on the solidified Muller Hinton agar plates with a sterile swab.

➤ The wells were cut in the agar using a well cutter aseptically.

➤ The kefir was centrifuged at 10000 rpm for 10 – 12 minutes for the separation of supernatant and the pellet.

➤ The supernatant ((150 µl) was poured in to the wells separately using sterile pipette.

➤ The pellet (150 µl) was poured in to the wells separately using sterile pipette.

➤ The plates were incubated at 37℃ for 24 hours.

Phase 4: Kefir with antibiotics against bacterial cultures for synergistic effect

➤ Pure bacterial cultures (*Staphylococcus aureus, Escherichia coli, Klebsiella sp., Pseudomonas sp., Bacillus subtilis., Proteus sp.,*) were isolated from their respective selective medium and sub cultured in a sterile Nutrient Broth and incubated at 37℃ for 6 hours.

➤ Muller Hinton agar was prepared and sterilized and poured on to the sterile petri plates.

➤ After 6 hours of incubation, the bacterial cultures was lawn cultured on the solidified Muller Hinton agar plates with a sterile swab.

➤ The wells were cut in the agar using a well cutter aseptically.

➤ The kefir was centrifuged at 10000 rpm for 10 – 12 minutes for the separation of supernatant and the pellet.

➤ 30% of antibiotics (tetracycline and vancomycin separately) and 70% of kefir's supernatent were mixed and poured in to the wells using a sterile pipette.

➤ 30% of antibiotics (tetracycline and vancomycin separately) and 70% of kefir's pellet was mixed and poured in to the wells using a sterile pipette.

➤ The plates were incubated at 37℃ for 24 hours.

Phase 5: Fractionation of bioactive compounds by thin layer chromotography

➤ Thin layer chromatography (TLC) was used to analyze the bioactive compounds (lactic acid, acetic acid, ethanol and bacteriocin) present in kefir.

➤ The extracts were spotted on to 10 cm×10 cm pre coated silica gel TLC plate. The solvent system used for this analysis comprised ofchloroform: glacial acetic acid : methanol: Water = 16:8:3:2

- The solvent system was saturated for half an hour prior to the experiment. The samples were spot inoculated on the TLC plate and after drying kept in the solvent system for separation. Following chromatography, the plate was stained with ninhydrinsolution (0.4g in 5 ml of acetone).
- After drying in hot air oven for 5 minutes, the Rf values were calculated and noted down.

$$R_f = \frac{\text{Distance from the baseline travelled by solute}}{\text{Distance from the baseline travelled by the solvent (Solvent Front)}}$$

RESULTS

Phase 1: Production of kefir (fig 1)
Kefir produced by following the Phase 1 methodology was used for the further examination

Phase 2: Treating bacterial culture with kefir
Following incubation the plates were observed for zone of clearance (Fig. 2A, 2B).

Phase 3: Treating kefir'ssupernatent and pellet seperately against bacterial cultures. The zones of clearance as observed for kefir pellet and supernatant against *Klebsiella* sp., *(3A), Pseudomonas* sp.,*(3B).*

Phase 4: Kefir with antibiotic against bacterial cultures for the synergistic effect The plates as observed for the synergistic effect of antibiotic 30% (a broad spectrum antibiotic Tetracycline and a narrow spectrum antibiotic Vancomycin) with 70% of kefir against *Staphylococcus aureus(4A), Klebsiellasp(4 B).*

The diameter of zone of clearance for the synergistic effect was tabulated. The obtained zones were compared with the standard Antibiotic susceptibility chart for 100% of antibiotics. The test results showed that S.aureus and Klebsiellasp were found to be sensitive when treated with 70% of tetracycline with 30% of kefir.

Phase 5: Fractionation of bioactive compound by thin layer chromotography (figure 5)
The R$_f$ value of the 3 fractionated bioactive compounds in kefir was tabulated(Table 3) .

DISCUSSION

The study was conducted to explore the antimicrobial activity of kefir and to find out the bioactive compounds responsible for this activity. Kefir was produced by inoculating kefir grains into the milk as done in the previous study quoted by Rattray and O'Connel [9] in the year 2011, where kefir grains were used as a natural starter culture during the production of kefir. The kefir was then treated with bacterial cultures namely *Bacillus subtilis, Staphylococcus aureus, Peudomonas sp, Klebsiella sp., Proteus sp.,* and *Echerichia coli* to determine its antibacterial activity by well diffusion method. In the previous study, the antimicrobial spectra of four types of kefirs (A, L, M, and S) fermented for 24, 36, 48, or 72 h were tested against eight food-borne pathogens namely *Bacillus cereus, Staphylococcus aureus, Listeria monocytogenes, Enterococcus faecalis, Escherichia coli, Salmonella Enteritidis, Pseudomonas aeruginosa,* and *Cronobacter sakazakii* by lawn culture method [10]. The kefir thus produced was then centrifuged for separating the supernatant and pellet. Both the supernatant and pellet were treated separately against the bacterial cultures in lawn. This was done to knowif the supernatant or the pellet had the antibacterial activity and the zone of clearance was observed to a visible level. The synergistic effect of kefir with antibiotics was studied by mixing 70% of kefir with 30% of antibiotics and treating against the bacterial cultures. The zones were observed and the diameter of the zones was compared with the standard antibiotic susceptibility chart. In the previous study, the alginate – coated probiotic was treated together with tobramycin against *MRSA* and *P. aerojinosa* which completely eradicated the pathogens [11].

To separate the bioactive compounds thin layer chromatography was done. The bioactive compounds present in the kefir are lactic acid, acetic acid, ethanol and bacteriocin. In the previous study, partial inhibition of B. cereus growth by lactic acid and acetic acid solutions at pH 3.5 and antimicrobial activity of ethyl alcohol solution produced by yeast were tested. Gaware et *al.*, [12] andMoraes et al., [13] reported the effect of a single unknown bioactive compound, bacteriocins or polysaccharides in controlling microbial growth. The presence of bacteriocinsmay be screened by pH neutralization, heat and enzyme treatment of the cell-free supernatants [14].Indira. M., etal[15] in the year 2015 reported that isolates showed maximum bacteriocin production for antimicrobial activity at 30- 50°C, beyond this temperature the activity was not found.

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Table 1: Synergistic effect of kefir with antibiotics

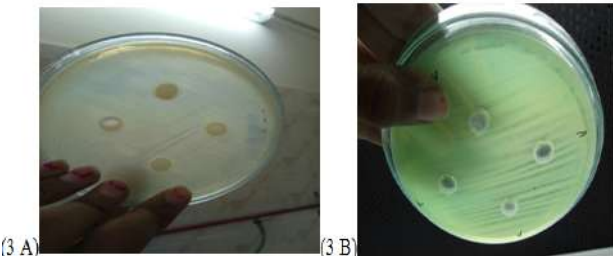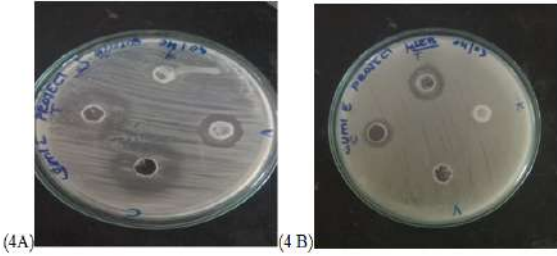| BACTERIAL CULTURE	TETRACYCLINE	VANCOMYCIN	CONTROL (TETRACYLINE)	KEFIR
S. aureus	2.2cm	1cm	1.5cm	-
*Klebsiella*sp,	1.2cm	0.9cm	0.7cm	-

Table 2:Interpretation of zone with antimicrobial susceptibility chart

ANTIBI OTICS	BACTERIAL CULTURE	% OF ANTIBIOTI CS USED	INTERPRETATIVE CRITERIA (mm for 100% antibiotics)			ZONE OF INHIBITION (mm)	CONTROL (TC) (mm)	FINAL INTERPRE TATION
			S	I	R			
TC	*S.aureus*	30%	22	19-21	19	22	15	S
	*Klebsiella*sp.,	30%	15	12-14	11	15	13	S

TC-Tetracycline S-Sensitive I-Intermediate R-Resistant

Table 3: Interpretation of R_f value in TLC of kefir:

MEASUREMENT OF THE COMPOUND TRAVELLED (SPOTS)	MEASUREMENT OF THE SOLVENT	R_f VALUE	STANDARD R f VALUE OF COMPONENETS
3.5cm	8.5cm	0.41	LACTIC ACID- 0.55
5.0cm	8.5cm	0.58	ACETIC ACID- 0.58
6.9cm	8.5cm	0.81	ETHANOL- 0.66

Fig. 1-Production of Kefir

Fig 2A – *E.coli***Fig 2B –** *Klebsiella*sp.

Fig. 3 Treating kefir'ssupernatent and pellet seperately against bacterial cultures. *Klebsiella* **sp.,** *(3A)*, *Pseudomonas* **sp.,***(3B)*.

Fig. 4.Kefir with antibiotic against bacterial cultures for the synergistic effect *Staphylococcus aureus(4A)*, *Klebsiella*sp*(4 B)*.

Fig. 5. Fractionation of Bioactive compounds by TLC

RESEARCH ARTICLE

Plant Disease Recognition Using Image Segmentation with Pixel Wise Classification Approach

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ABSTRACT

In the cutting edge scenario, image processing has a lot of applications in various fields such as medical, agricultural, industry, etc. The main step in image processing is image segmentation, which extracts the objects and information present in the image. Detection of disease in plants by hand in an isolated locality is a wearisome job. So, automatic discovery of plant leaf diseases is necessary in agricultural fields to improve productivity. This work focuses on extracting diseased and healthy segments of the image using a pixel-wise classification approach. This work builds a model to segment the image that automatically detects the different segments of the leaf, such as healthy and diseased parts. The performance measurement shows that the proposed pixel-wise classification gives better performance among threshold and region based technique. To recognise the disease in a leaf, an SVM classifier is used. This gives better classification accuracy as compared to other classification algorithms with global colour and GLCM features.

Keywords: Pixel wise Classification, Image segmentation, Clustering, Classification.

INTRODUCTION

Nowadays, significant number of images are produced using various imaging modalities in various fields such as medicine, remote sensing, agriculture, etc. Plants are one of the most effective resource to prevent global warming. The paddy crops, which cater to maximum food necessities, undergo a large number of diseases caused by environmental conditions and are malnourished. This results in a huge loss for the farmers, which may be reduced

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with the advice of the experts [2]. But remote locations may face the same consequence because of the location constraint. Technological solutions becomes a good answer for all these problems, as they would not require travel. Without having to travel to an avid location, professionals can check the condition of the crops and provide legible advice via some technological device [1].Rice is the staple food in India, and our economy would become crippled without the production of crops. Mechanical, nutritional deficiency, climate condition, and genetic disorder all have an impact on the growth of paddy crops. It is noticed that microbes and microbes cause the highest impact on their growth [11]. The disease in paddy crops is one among the most important causes of loss in Arkansas rice production. Diseases cause an annual loss of 8 to 10%.Chemicals and other disease control methods lead to a rise in the rate of production [3]. Environmental hazards lead to the outbreak of diseases which are harmful to the crops and can be acknowledged by the variation in the plants [8] [10]. Recently, many paddy diseases have been identified. According to the conventional process of detecting diseases manually by farmers, there are often mistakes [4] [9]. This takes more time-consuming and difficult to implement. Moreover, the accuracy level is also seen to be very low, hence an advanced technology for the identification and eradication of the disease is highly advisable. In order to solve this problem, "image processing" is the distinguished solution. This work elaborates on the perception of developing some process for the early detection of disease in plants. This technique detects only the portion of the leaf that is damaged.

Segmentation plays an active role in recognising whether the leaf has disease or not. Many segmentation techniques have been proposed during the last few decades. The primary purpose of Image segmentation is the domain-independent dividing of an image into a collection of disjoint regions that are visually dissimilar and meaningful in relation to grey value, texture, or colour features. If the process of segmentation is carried out correctly, all the other steps for analysing the image becomes easier. The segmentation of an image can be accomplished by a discontinuity detection (edge-based), a similarity detection (region-based), and pixel classification approach.

In the region-based approach, the pixels in the image is organised into groups, gathered on their similarity. In the edge-based approach, pixels are composed in to groups with their abrupt fluctuations in the intensity value. Ideally, edges produce an outline scene present in the image. From the edges, vital features corners, lines, and curves can be mined. Furthermore, this extracted data is used for higher-level image processing algorithms (e.g., recognition).The edge detection process filters out less relevant information and preserves important structural properties of an image. Pixel classification approach labels each and every pixel in the image. It utilises both clustering and threshold technique for labelling the pixels. A classification procedure is carried out after extracting features from the image. Classification is the method of grouping the input data into a different classes. Image features at the pixel level are recovered. Next, feature vectors are produced from them. Pixels are labelled using classification techniques with these feature vectors. This work find a segmented model which categorise the pixels in the leaf image as healthy, diseased, or background with better classification accuracy than the existing segmentation techniques. Here, image segmentation can be considered as a pixel classification problem.

Related Works

Many studies have been conducted to identify, recognize, and classify diseases in images of plant leaves. For recognizing the disease, most of the studies focuses on three key phases image segmentation, feature extraction, and classification of disease. Various algorithms are utilized to take out the affected area of the leaf, then it is used for higher level study. This part gives a review of image segmentation algorithms for detecting and classifying plant diseases. Hiary et al[5] propose a software solution that uses K-Means Clustering to find out and classify plant diseases in leaves automatically. Valliammal et al[6] used fuzzy based thresholding and clustering to eliminate the complicated background and retrieve the leaf image. Faith praise Fina et al[7] suggests K-Means clustering for automatic plant pest detection & recognition.

Suman *et al* [12] developed a model to detect bacterial leaf blight, brown spot, narrow brown spot, and rice blast diseases using 8-connected component analysis for disease part segmentation. Plant leaf diseases automatically

detected by using GA based FCM is proposed by [13][16]. Joshi et al[17] suggest a method to extract the diseased part from the leaf with YCBCR Color space.K-Means Clustering is employed to locate illness spots in [18].Meenakshi *et al* [19] utilised K-means clustering or 3 means clustering for disease detection in plants. Ali *et al* [20] suggested FCMmethod for detecting plant leaves. Delta E colour difference algorithm segments the concerned parts from the Leaf is suggested by [21]. Kao *et al* [22] proposed Convolutional Auto Encoder with clever edge detection to find the disease afflicted part for lycopersicon identification and interpretation into three maturity levels.Zhang *et al* [23] proposed a hybrid clustering approach constructed on Super Pixel Clustering and the Expectation Maximization (EM) algorithm for segmentation of leaf image.For rice disease classification and identification, diseased suffering parts are segmented from the leaf using threshold in HUE and Saturated Component..

PROPOSED METHODOLOGY

This methodology proposes a method for labelling the pixels present in various portions of the plant leaf for detection of disease. The proposed flow used as follows A) Image Pre-processing (B) Image Segmentation (C) Feature Extraction (D) Recognition of Disease.

Image Pre-processing

Pre-processing is accomplished to rise the reliability of the optical inspection. Filter operations help to bring about the image more detailed and aid in quick evaluation. Cropping, rotating, normalizing, contrast enhancement, and filtering help the user to improve the visual excellence of an image. CNN is used for Nosie Removal. The image enhancement pre-processing step improves the image's visual quality. It denotes the process of making the display more suitable by adjusting gray value of digital images. Image enhancement is done with contrast enhancement technique.

Image Segmentation

This assists to provide a better granular understanding of an image through the formation of pixel-wise mask for the objects. Clustering and thresholding are employed for categorising the image pixels. Image segmentation is the technique to categorise each pixel in an image, allowing it to be subdivided with different color, texture, and edge or shape information Generally, image segmentation is performed on similarity-based and discontinuity-based and Pixel-Wise Classification Approach. Pixel-wise classification is performed in three levels. The first stage involves pixel wise labelling with the purpose of building a segmentation model. It's identified as the labelling phase, and it's where the data matrix and label are created. Supervised methods [Classification Tree, SVM, and Adaboost] are used at the second level for learning to predict the model with the knowledge acquired from the labelling phase. At the second level, an image segmentation model is constructed from the information found from the labelling phase. In the third level, prediction phase categorise the fresh data and assign a class label to it. FirstPixel level colour and edge features are taken out from the image. Then the extracted features are automatically labelled into background pixels, healthy and diseased pixels using an Improved Fuzzy C Means Clustering based Threshold. The general flow of the steps are followed in this research work to detect the affected portion of the leaf is given below.

Improved Plant Disease Detection Algorithm (IPDDA)
Step 1: Input leafimage taken by a digital camera.
Step 2: Convert the image to the YCBCR colour space from the RGB colour system.
Step 3: Categorize the pixels in the image using an Improved Fuzzy Clustering-based Threshold Technique.
Step 4: Tagging every pixel with the label and training using the SVM approach
Step 5: Generate segmented images from the original image by the predicted value.
Step 6: Divide the infected and uninfected portions of the plant leaf automatically.

Improved Fuzzy C Means Clustering Based Threshold (IFCM)

Fuzzy C means (FCM) clustering is an unsupervised machine algorithm that is widely used in many applications. The FCM algorithm partitions the pixels of an image into k groups. Pixels in each group have similar colours or other low-level features.

IFCM Algorithm

'D' denotes the number of features.

Dataset 'X' is a collection of pixel-level colour and edge features extracted from the input image.

K is the number of groups.

'n' size of the dataset.

Input: X, D, K fuzzification value m > 1.

Output: K numbers of segmented images

Procedure:

Step 1: Define a membership matrix M of size $n \times K$

Step 2: Assign K seed points arbitrarily contained by the range of (0-255). Let the seed points be $c_1, c_2, \ldots \ldots, c_k$

Step 3: calculate the distance measure $d_{ij} = \|x_i - c_j\|$ using Euclidean distance, for all seed points c_j, j=1,2,...,K and data objects x_i, i=1,2,...,n.

Step 4: compute the fuzzy membership matrix M_{ij}

$$\underset{1 \le i \le n,\, 1 \le j \le K}{\forall} U_{ij} = \begin{cases} \dfrac{(d_{ij})^{\frac{1}{1-m}}}{\sum_{i=1}^{K}(d_{ij})^{\frac{1}{1-m}}} & \\ \begin{cases} 0, & j \ne I_i \\ \sum_{j \in I} M_{ij} = 1, j \in I_i, & I_i \ne \emptyset \end{cases} \end{cases}$$

(1)

Where $I_{\forall \le i \le n}\, i = \{j | 1 \le j \le K;\, d_{ij} = 0\}$

Step 5: compute new seed points C_j

$$\underset{i \le j \le K}{\forall} C_j = \frac{\sum_{i=1}^{n}(M_{ij})^m x_i}{\sum_{i=1}^{n}(M_{ij})^m}$$

(2)

Step 6: Repeat steps 3 to 5 until convergence

Step 7: Assign labels to data objects according to the membership value.

Step 8: Group the Corresponding Pixel and create an image in YCBCR Color.

Step 9: Find the threshold automatically using histogram of the segmented image.

Step 10: Obtain RGB segmented Image.

Edge Detection

Edges produce an outline sketch of information present in the image, and edges play a vital part in segmentation. Edges denotes the boundary between the objects and also contain facts about the shape of the object. This information can be used in further analysis of an image to get higher level details. Roberts', Sobel, Prewitt, Kirsh, Robinson, Marr-Hildreth, LoG, and Canny Edge Detection are the edge detection methods used for detecting the outline shape or boundary of the image. Canny edge detection is used to discover the edges and extract shape information from them. The algorithmic steps are as follows:

- Convolve Gaussian function with input image to obtain a smooth image.
- Apply the Sobel operator
- Apply non-maximal.
- Apply a threshold to find the edges.

Support Vector Machine Classifier

Support Vector Machines is a supervised machine learning method. It is employed in the categorization and regression of data. Vapnik (1982, 1995) and Chervonenkis (1974) proposed this robust prediction method constructed on statistical learning frameworks. In SVM, different classes are in a hyper plane with multidimensional space. The SVM generates the hyper plane in an iterative manner to minimise the error. In SVM, the datasets are classified into classes with a maximum marginal hyper plane (MMH). The main goal of SVM is to design a hyper plane that classifies all the training vectors into different classes.The hyper plane is drawn with the widest possible distance between the two points, called support vectors. Now, pixel-level features such as colour and edge are used to classify and identify anthe pixel using an SVM classifier. Segmentation of different portions of the leaf is done with the following steps:

Step 1: Load all the parameters attained from the above stages for training with labels.
Step 2: Insert the test image features into a variable called "test".
Step 3: Use an SVM classifier to predict the test features of an image with a training dataset.
Step 4: Visualize the diseased part of the leaf image.

Feature Extraction

It's acrucial step, and the correctness of a classification procedure is influenced by the collection of features. Extracting appropriate features is essentially dimensionality reduction, which successfully denote the remarkable portions of an unhealthy region in a compact form. Outline, shade, and surface-based attributes signify different leaf-based classification attributes. One important factor in determining whether a leaf is healthy or unhealthy is its shape. The shade of the contaminated area is differs from the healthy area. Likewise, the texture of the infected area can establish the nature of the problem and can be used as a major forecaster. GLCM, Color features are extracted from the image globally and are described respectively.

Color

In an image, colour information is represented by different colour models. Color space is a mathematical model for representing colour information as three or four different colour components. Different colour models are used for different applications, such as computer graphics, image processing, television broadcasting, and computer vision [23]. Various colour spaces are available for processing colour images. These are RGB-based colour model (RGB, normalised RGB), hue-based colour space (HSI, HSV, and HSL), luminance-based colour space (YCBCR, YIQ, and YUV), and perceptual colour space (CIEXYZ, CIELAB, and CIELUV) [14]. The RGB colour space is not preferred for color-based detection and colour analysis because of the mixing of colour (chrominance) and intensity (luminance) information and its non-uniform properties. Efficient separation of colour and intensity information and transformation is easy in the YCBCR colour space compared to HSI or HSV [14]. HSV (Hue, Saturation, Value), also known as HSB (Hue, Saturation, Lightness), is commonly used by artists because it's often more natural to think of a colour in terms of hue and saturation than in terms of additive or subtractive colour components [25]. Here statistical information about red, green, and blue components is used as global features.

Gray Level Co-occurrence Matrix (GLCM)

The surface is the more significant portion for disease identification as it signifies more evidence with the unhealthy region. The shade evidence is previously prearranged by with the help of colour instants.. The GLCM is considered over grayscale input image that is resampled into 8 gray-levels. It estimates how often the pixel gray-level value is positioned together with pixel i and j. For an image with 8 different gray-level concentrations, the GLCM G of measurement 8×8 is definite over $m{\times}n$ image I, with an equalizer parameter $(\Delta a, \Delta b)$ formula in eq. 1 terms it.

$G\Delta a, \Delta b(x,y)=$

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$$\sum_{a=1}^{m} \sum_{b=1}^{n} \begin{Bmatrix} 1, \text{if } I(a,b) = x \text{ and } I(a + \Delta a, b + \Delta b) = y \\ 0, \qquad\qquad\qquad\qquad \text{otherwise} \end{Bmatrix}$$

(3)

x, y indicates the intensity value of pixels
a,b denotes the locations in the matrix I of an image
$\Delta a, \Delta b$ are offset values that denotes the spatial relation in the matrix
I(a,b) – denotes pixel values at position (a,b).

Disease Recognition
The dataset is built from diseased portion of leaf in order to recognise the disease existing in the plant. Several features from the image are extracted. The information is then saved in a vector format. SVM classifier is used for recognizing the disease. The proposed method provides greater accuracy with SVM classifier to recognise diseases caused by pathogens.

RESULTS AND DISCUSSIONS

This dataset was built from some random images in the internet. The test images consisted of images with various luminance conditions and with various backgrounds. The image formats suitable to the algorithm are jpeg, png, bmp, etc. The algorithm run on Intel® Core TM i5 CPU M 430 running at 2.27 GHz with 4.00 GB of memory. With proposed IPDDA, disease-affected portions of the leaf are segmented. The features extracted from the segmented image are colour and GLCM. The extracted features are given as an input to the SVM to recognise the disease. The SVM classifier gives a better recognition rate as compared to KNN. Accuracy parameters is used as a measure to determine the success of the work in two stages, such as segmentation and recognition. The accuracy of pixel classification/segmentation is compared in Table 1 to existing threshold and region-growing method. The classification accuracy is displayed in Table 2.Using colour and GLCM characteristics with a KNN and SVM classifier. The accuracy of SVM classifier is higher as compared to KNN.

CONCLUSION

Image processing contribute a significant part in agricultural field. An image segmentation approach is employed to recognise the leaf in a plant has disease or not. In the pre-processing stage, the CNN denoising technique is utilised to remove noise from the image. Various image processing techniques, such as feature extraction, clustering, classification, and thres holding are used to create the suggested image segmentation model. When compared to previous approaches such as threshold and region growing based segmentation, the suggested image segmentation model using Improved Plant Disease Detection Algorithm (IPDDA) provides greater accuracy.The IPDDA algorithm's segmented output is fed into an SVM classifier, which recognises the disease using global colour and texture features. With comparison to KNN, the SVM classifier has higher accuracy. In the future, image segmentation with hybrid feature extractor can boost recognition accuracy.

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Table 1 Pixel Classification/Segmentation Accuracy

Technique	Pixel Accuracy
TBS	93
RBS	94
IPDDA	96.4

Table 2 Classification Accuracy

Features	Classifier Algorithm	
	KNN	SVM
COLOR	78	83
GLCM	72	77.8
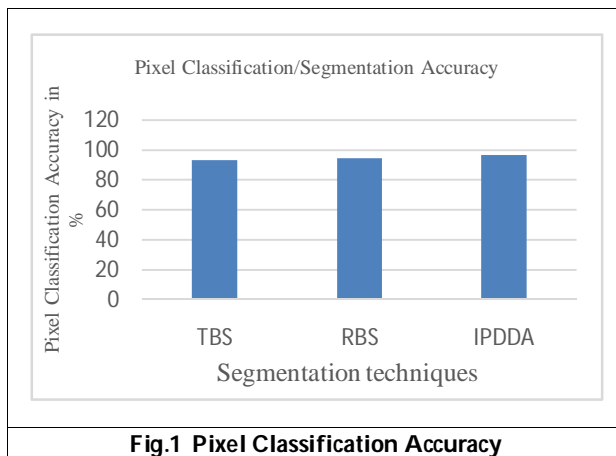
Fig.1 Pixel Classification Accuracy	**Fig.2 Classification Accuracy**
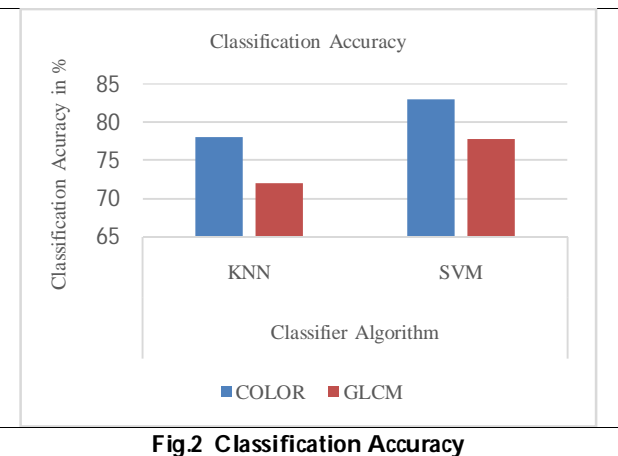
40470

RESEARCH ARTICLE

Python Libraries for Computer Vision - A Comparative Study

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ABSTRACT

Computer Vision helps us to obtain a high level understanding from images and videos. It automates the process of the human visual system which understands an image or video. Python provides a variety of libraries for the purpose of computer vision. These libraries contain traditional image processing as well as modern computer vision functions for feature computation from images and videos. The main libraries are scikit image, open CV, pillows, simple Tk, script, pillow etc. This paper provides a comparative study of these libraries. It also provides the different commands used for each operation by different libraries. This includes commands for Morphing, color spaces conversations, Histogram equalization, edge detection, image contour etc.

Keywords: Python, OpenCV, Imutils,

INTRODUCTION

Python programming language is a general-purpose language that uses a significant indentation in its design which highlights the code readability[1]. It uses an object-oriented approach for implementation. This helps to write logically clear code for the project by both new and experienced programmers. This programming language helps in a variety of fields like Data Science, computer vision, machine learning, etc. because of its vast community support, free and open source. This language provides a variety of libraries that makes complex computation very easy and allow developers to carry out visualization-based tasks at high speeds[2]. Computer Vision helps to understand images or videos. Computers can derive meaningful information from digital images, videos and other visual inputs by implementing a computer vision algorithm, this information helps to make recommendations or take actions. Computer vision enables the system to see, observe and understand whereas Artificial Intelligence makes computers

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think.[3] With the advancement of artificial intelligence and revolution in the field of deep learning and neural networks, the field of computer vision has been able to surpass humans in some tasks related to detecting and labelling objects[3]. The availability of a large amount of data and its use for training makes computer vision better.[3]

Python Libraries for Computer Vision

Python is a general purpose programming language. It works well with windows Linux, raspberry pi etc. its syntax are very easy and similar to English language and in fewer lines[4]. The programs can written either as functions, procedures or object orient way. Python provides a variety of libraries for computer vision[5][6]. The common libraries are

OPEN CV

Open CV (Open Source Computer Vision Library) is an open-source computer vision library,.Intel was created and released in 2000. Using 2500 + classic and state of art of algorithms helps to provide a common infrastructure to computer vision applications[7][8]. Red-eye removal, Face detection and recognition, extraction of 3D models of objects, object identification,, tracking moving objects, stitching multiple frames together into a high-resolution image etc are made easy because of these algorithms[6][9].

OpenCV is used for
- Reading and writing images
- Save and Capture videos[10]
- Process images (filter, transform)[10]
- Perform feature detection[11]
- Detect specific objects such as faces, eyes, cars, in the videos or images.[12]
- Analyze the video, i.e., estimate the motion in it, subtract the background, and track objects in it[13].
- The main OpenCV library modules are
- Core Functionality- which contains data structures and storing images multidimensional array Mat.
- Image Processing- contains various image processing operations such as image filtering, geometrical image transformations, colour space conversion, histograms, etc.[14][11]
- Video contains various video analysis methods. This includes motion estimation, background subtraction, object tracking etc. [7]
- Video I/O - contains the video capturing and video codecs
- Calib3d contains basic multiple-view geometry algorithms, single and stereo camera calibration, object pose estimation etc.[11]
- Features2d- contains the concepts of feature detection and description
- Objdetect - contains the detection of objects and instances of the predefined classes such as faces, eyes, mugs, people, cars etc[15]
- Highgui- provides UI capabilities and easy-to-use interface
- Camera calibration and 3D reconstruction (calib3d module) - extract 3D world information from 2D images[11]
- Deep Neural Networks (dnn module) - infer neural networks using built-in dnn module
- Graph API (gapi module) - graph-based approach to computer vision algorithms building
- GPU-Accelerated Computer Vision (Cuda module) - utilizing the power of video card to run CV algorithms

Tensor Flow

Tensor Flow is a free open-source platform [16]. It provides tools, libraries, and resources [17] for Artificial Intelligence which includes Computer Vision. In 2015, the Google Brain team was created to build and train Machine Learning models related to computer vision that include facial recognition, object identification, etc.[18] For mobile devices, Google released the Pixel Visual Core (PVC) in 2017. The main modules are
- Audio module: used to encode and decode WAV files..

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- autograph module: Eager-style Python files are converted into TensorFlow graph code.[19]
- Bitwise module: Operations for manipulating the binary representations of integers.
- Compact module: Compatibility functions.
- Data module: used to import datasets.
- graph_util module: a tensor graph in python is manipulated
- Math module: Math Operations [18].
- Sets module: Tensor Flow set operations [18].
- Signal module: Signal processing operations [17].
- Strings module: string operations.

Simple CV

Simple CV is an open-source computer vision framework used for building computer vision applications[20]. Simple CV is simple as its framework, gaining access to several high-powered [6] computer vision libraries such as Open CV without learning all the CV concepts in-depth such as file formats, colour spaces, buffer management, or file formats[19]. Simple CV allows experimenting in computer vision using images or video streams. It can be from webcams, FireWire, mobile phones, Kinects, etc.[21]. This framework for performing quick prototyping. The common modules are

- Simple CV. Camera module- manages Camera access
- Simple CV. Color module -used to model the colour of foreground and background objects by using a training set of images
- Simple CV. Display module used to write images[19]
- Simple CV. Drawing Layer module- provides a way to mark up Image classes without changing the image data itself. It is also used to perform the geometric operations[20]
- Simple CV. Font module- allows you to create a font object to be used in drawing or writing to images.[21]
- Simple CV.I mage Class module- used to encapsulate the colour space of a given image.[20]
- Simple CV. Stream module- allows the user to stream a jpeg encoded file to an HTTP port. [22]

D SCIKIT-Image

Scikit-Image is an open-source Python library that is used for image processing. It is basically an image processing toolbox for SciPy. The library provides a variety set of image processing routines[23]. This image processing library provides a well-documented API and uses it in research, education and industry applications[24]. The main modules are

- Input/output, data types and colorspaces
- Image preprocessing / enhancement
- Local filters and Non-local filters
- Edges and lines- include finding contour, convex hull, ridge operators etc.[25]
- Geometrical transformations and registration[26]
- Mathematical morphology
- Image segmentation includes Binary segmentation: foreground + background, Marker-based methods[24]
- Measuring regions' properties
- Data visualization and interaction[23]
- Feature extraction for computer vision

E Pillow-PIL

Pillow is also known as PIL is a Python Imaging Library which supports image processing tools. This library is good for image archives and batch processing.[27]It support a number of file formats like TIFF, BMPetc. The main Functionality are[28]

- The image module helps to Displaying the image, Getting the mode (color mode) Getting the size of the image, Rotating an image etc.

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- The Image Draw module support 2D graphics for Image objects and to create new images, annotate or retouch existing images etc.[27]
- The Image File module main features are to support open and save an image.
- The Image Grab module copy contents to PIL image memory from the screen or the clipboard.[27]
- The Image Path module process 2-dimensional vector data. [28]
- The Image Color module contains color tables and converters from CSS3-style color specifiers to RGB tuples
- This method is used to create a thumbnail of an image using draft([28]
- To merge function helps to merge two or more images[29]
- Crop, filp, blur. Rotate are very easy to implement using pillow[27]

Imutils

Imutils is a computer vision package. It contains convenience functions such as translation, rotation, resizing, skeletonisation, displaying Matplotlib images, sorting contours, detecting edges, among others quite easy[30]. The main modules are

- Open CV functions can be found by function name[31]
- Translation: Translation is the shifting of an image in either the x or y-direction.[32][33][34]
- Rotation: Rotating an image for a given angle[30]
- Resizing: Resizing an image by maintaining the aspect ratio [31]
- Skeletonization: The process of constructing the "topological skeleton" of an object in an image.[30][35]
- URL to Image: the URL of the image to download and convert to a NumPy array in OpenCV format.[35]
- Automatic Canny Edge Detection: This uses the median of the grayscale pixel intensities to derive the upper and lower thresholds [31].
- 4-point Perspective Transform: Helps to obtain a top-down, "birds-eye view" of the ROI.
- Sorting Contours: sort a list of contours from left-to-right, right-to-left, top-to-bottom, and bottom-to-top, respectively [31].

Deep Face – Free Deep Learning Library for Face Recognition

Deep Face is an open-source computer vision library that is used for facial recognition. This helps to find facial attribute analysis (age, gender, emotion and race)[36]. This library is licensed under the MIT License and permit the developers to modify the library, use the library and distribute the library. [37] This also helps to perform,

- Face Verification: comparing a face with another and verifying that it is a match or not. [38]
- Face Recognition: finding a face in an image database. [32]
- Facial Attribute Analysis: describing the visual properties of face images. [33]
- Real-Time Face Analysis: for the real-time video feed from the webcam it tests face recognition and analysis facial attributes [32].

YOLO – Object Detection

YOLO or You only look once! Is a real-time object detection system. It was created by Joseph Redmon and Ali Farhadi from the University of Washington in 201[39]. It is extremely fast and accurate as compared to the other object detectors because it applies a neural network to the full image in order to classify the objects [35][40]. It looks at the whole image so its predictions are informed by a holistic context of the image.
The files needed are[39]

- yolov3.cfg – The standard config file/ directory.[35]
- Yolo-tiny. cfg – cfg/ directory will be optimized[35].
- yolov3.weights – Pre-trained weights file available in the darknet/ directory.[35]
- Yolo-tiny. weights – optimized weight file available in the darknet directory[41]
- coco. names – item list that model can recognize[35]
- coco. data – A config data file kept in the cfg/ directory[40]

CONCLUSION

Computer vision is a new research area where a lot of libraries are developed day by day. The updates provided by the different community is also high. So selecting a good library is a very difficult task. We can use a different library for our project and knowledge about the different libraries is good for those who work in this field. All these libraries can make your workflow easier with their simpler implementation for specific functions[35]. The future development of image processing is mainly based on python and these libraries help to a great extent.

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Table 1 . Comparison : **The following table shows the advantage and disadvantages of different libraries**

Library	pros	cons
Open CV	Open Source and free Usage Community support and access to 2,500 + algorithms	Compare to MATLAB it is difficult to use
Tensor Flow	open-source platform and multiple languages compatibility features are updated constantly[9]	an extremely resource-hungry toolkit[9]
Simple CV	Optimized algorithm and free to use good documentation	Only python programming language support
Deep face	Open-source and Free for commercial use easy to install, Lightweight and supports the popular models and detectors Optimized to perform real-time on-device inference (Edge AI)[9]	Cloud API is not available
YOLO	Exceptionally fast, highly accurate, and background errors are minimal The algorithm has top-notch learning capabilities[9]	To detect small objects is not effective and limited community support
Pillow	Have more functionality. Runs in all operating system and support variety of image formats	Not work well with advanced image processing no option of disable delta frame for animated gif

REVIEW ARTICLE

Contemporary Factors Responsible for Poor Natural Regeneration in *Shorea robusta* and its Management: An Overview.

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ABSTRACT

The forest system has been persistently affected by the destructive anthropogenic activities, natural regeneration problems along with indiscriminant use of forest resources especially in case of tropical moist deciduous forests. *Shorea robusta* is one of the prime species of central Indian tropical moist deciduous forests. There has been an alarming decline in the *Shorea robusta* forest biodiversity owing to illegal felling, forest area encroachments, illegal wildlife poaching and ever expanding commercial fuel wood plantations in the forest area. In its highly diverse and affluent plant bio – ecosystem Sal has a special status with respect to commercial, ecological as well as medicinal importance and also supports a variety of other medicinal plants in its vicinity. Natural regeneration of *Shorea robusta* has been associated with a large number of intrinsic as well as extrinsic factors. Intrinsic factors like dieback syndrome, severe heartwood decay, blight, Sal borer attack and extrinsic factors like forest fires, over – exploitation, grazing etc. have been identified as serious issues to be taken care of. This review tends to provide a comprehensive understanding of the factors responsible for rapid depletion of Sal forests and to find out appropriate and effective scientific concept based advance silviculture management strategies, reported in the literature, which are required to be applied in lieu of Sal forest conservation. An in-depth assessment of potential shortcomings and climatic stress exerted during natural regeneration along with other anthropogenic factors is the need of the hour prior to strategy formulation in context to conservation of Sal forests.

Keywords: *Shorea robusta*, Genetic diversity, Natural regeneration, Climatic Variation.

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INTRODUCTION

The contemporary global shift in the climatic conditions has a direct bearing on the processes pertaining to efficient regeneration of different species. The process of genesis for the survival and perpetual of trees and forests as a whole is generally referred to as the regeneration process (Bhuyan et al. 2003). The *in-situ* seed germination from the pre – existing genetic material available, forms the basis of the natural regeneration of *Shorea robusta* and is responsible for its fortification and restocking. The key factors that determine the particular forest species natural regeneration pattern depends on species (Adhikari et.al. 2017). The regenerative behavior of the forest is depicted by the vertical stratification of various species while the regenerative potential is indicated in the occurrence of seedling and saplings of the species present in the vertical strata of the forest. Gregarious occurrence of Sal Forest has been reported in the southern Himalayas with its distribution in India, Nepal, Bangladesh (Orwa et al. 2009) along with some parts of South China and Khanabarti river in Bhutan (Gyaltshen et al. 2014). The area under Sal forest cover in various countries like Bangladesh (0.12 million hectare), Nepal (1.4 million hectare) and India (0.57 million hectare) has been reported by various authors (Alam et al. 2008, Rautiainen 1999, Rathore 2000). In Indian context, Sal forest cover can be categorized on the basis of its distribution in four major regions. In the northwestern region, Sal forest cover occurs in the lower belt of Himalayan region in Kangra valley of Himachal Pradesh along with the northwestern parts of Uttrakhand. In the north central region, it is distributed in the states of Uttar Pradesh, Bihar, Jharkhand and foothills of northwest Bengal. In the north eastern Himalayan region, Sal cover extends in the Assam valley including Meghalaya and Tripura. In the central region of the country it has its spread in the states of Madhya Pradesh, Chhattisgarh and Orissa (Orwa et al. 2009, Adhikari 2017). As reported by the ISFR 2015 the total geographical area covered by the Sal forests in the Shiwaliks, Gangetic Plains and Central Indian plateau are about 13.6%, 2.2% and 8.9% respectively.

Shorea robusta, commonly known as Sal, is considered to be one of the most important tree species in terms of socio – economic aspect. It is among the prime category of tree species due to its dual properties. It is commercially important timber species and is utilized as a source of construction wood, tannin, gum, oil, fodder and fuel wood and as a medicinal tree owing to its pharmacological utility as an analgesic, antipyretic, anti – inflammatory and anti - oxidant(Soni et al. 2013). Its medicinal properties and uses have been documented in traditional practices since ancient times (Agarwal et al. 2018).It is also used in sleepers, ship building and other commercial uses. Thus, it requires proper management strategies for its conservation in order to have a sustainable supply of Sal. A good seed year along with In time commencement of monsoon are required for proper seed germination and its natural regeneration. However, certain factors collectively have been identified to be the causing serious damage to the overall Sal cover. Climatic and edaphic factors have the major role in controlling the distribution pattern of Sal forests. In Indian context, the major distribution lies in the plains, lower foothills of Himalayas including the valleys. Majority of the Sal forests are undergoing through a regeneration shift due to increase in temperature, precipitation and less moisture availability in the natural forest. Contemporary requirement is to assess the most likely effects of the predicted climate shift on the forest ecosystems at species level in particular. Occurrence of seedlings, saplings and trees ranging from young to old age is a representative of regenerating forest species (Chauhan et al. 2001).

This distribution range provides specific information pertaining to regeneration potential of a species. Maximum numbers of individuals of a species present in the seedling stage in the distribution curve and a subsequent slope at the next stage is termed a reverse J shaped curve and is representative of a good regeneration potential of the *Shorea robusta*. Another possibility is a bell shaped curve which represents the area with even aged population which has been managed under some conservation system. The J shaped curve can also be formed in an old grown forest population and shows unsuccessful regeneration process (Agarwal et al. 2018). Since the last few decades there has been great anthropogenic pressure on Sal forest cover and Indiscriminate deforestation has led to alterations in the dynamics of Sal cover (Chaitale and Bahera 2012). The prime hindrance in maximization of Sal forests for its sustainable use has been the problem of regeneration. Studies conducted on Sal forest structure, diversity and regeneration, are confined to smaller zones which shows lacunae in silviculture management.

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Background

According to the available climatic conditions Sal can be both deciduous (in dry conditions) as well as evergreen (in wet climate). It is a subtropical species with higher light requirements and the leaves are never shed completely at the same time. Ideally, natural Sal stands occur within the latitude range of 20 – 32^0 North and longitude range between 75 – 95^0 East (Gautam and Devoe 2006). The range of elevation lies between 100 – 1500 meters (Pandiyal 2012). It is characteristically slow growing species and the fully mature trees may reach up to the height of 30 – 50 meters (Chitale and Bahera 2012). The process of natural regeneration is highly dynamic and can be conspicuously categorized into two different steps namely the recruitment phase and the establishment phase (Puri, 1960). The problem in this process occurs at the recruitment phase pertaining to die back phenomenon according to (Rautiainan and Suoheimo, 1997, Mishra et. al. 2020. This syndrome causes discrepancy in the phase of seedling to sapling conversation. A living root system of *Shorea robusta* grows continuously in a periodic fashion until an enhanced root stock system is formed. In case where the seedlings overcome the die – back process; tend to form 'large and whippy' or 'small and woody' pattern. Apart from that, the process of natural regeneration is further associated with a large number of intrinsic and extrinsic factors which are responsible for lower viability like soil properties, structure of the canopy, availability of sunlight, low seed viability, competitive interaction with other species for space and nutrients. And the major root cause of these instabilities is rapid urbanization and climatic stress (Adhikari et al. 2017). Kumar 2018 reported that the climatic shift has been observed to be the main cause of early flowering and reduced seed setting in the Uttrakhand state forest, thus disturbing the natural regeneration process of Sal cover. Additionally, the combination of climatic disturbances along with frequent forest fires proves to be highly bizarre for the vigor and susceptibility towards disease defense of Sal shoots (Ganguly, 2019).

Ecology and Phenology of *Shorea* Species

Shorea robusta belongs to the family Dipterocarpaceae which further divided into 3 sub – families covering17 genera and 511 species in total (Ashton, 1982). It is a hermaphrodite species. It is the state tree of two states namely Chhattisgarh and Jharkhand, India. Thysanoptera are insect group pollinate this tree while exclusive pollen vector for *Shorea robusta* is wind (Momose *et al.* 1998). Ovoid seeds with nearly 8mm diameter and 2gm weight are characterized by two short and three long wings (Jackson, 1994) to facilitate wind pollination. It is a biannually fruiting plant after attaining maturation at the age of 15 years (Orwa *et al* 2009). However seeds are produced year after year with good seed year after every 3 – 5 years. Species belonging to different strata of Sal forest in Bhabar/Teari and Hilly terrain were documented by Stainton (1972) while Rana et al (1988) enlisted the species that dominate Sal forest belonging to different age groups and thus are representative of the same. The top canopy in Bhabar/ Terai region were predominated by *Shorea robusta, Terminalia tomentosa, T. belerica, T. chebula, Adina cardifolia, Anogeissus latifolia, Lannea grandis, Scleichera trijuga, Syzygium cumini* while that in the lower canopy is a bit different and is constituted by major species like *Mallotus philippinensis, Semicarpus anacardium, Dillenia pentagyna, Kydia calycina, Apotosadioca, Casearia tomentosa, Buchnania latifolia*. At the shrub level , the canopy of the forest cover in Bhabar/ Terai region the species observed were entirely different with species like *Ardisia humilis, Zizyphus rugosa, Clausena spp., Barleria cristata* and those at the lianas canopy level were (*Spatholobus roxburghii, Bauhinia vahlii*) similar to that of the Hilly region. The shrub level of the Hilly terrain consisted of species like *Hamiltonia suaveolens, Phoenix humilis, Indigofera pulchella, Flemingia strobiliferra* and the lower canopy was characterized by *Nyctanthes arbor tristis, Kydia calycina, Leucomeris spectabilis, Glochidion velutinum, Symplocos racemosa*. Species of the top level were reported to be an admix of species like *Shorea robusta, Lagerstroemia parviflora, Anogeissus latifolia,Adina cardifolia, Semicarpus anacardium, Bauhinia variegata, Dillenia pentagyna, Buchnania latifolia*.

Similarly, the species when observed at different age levels showed variation at different strata. The species of the tree layer (*Shorea robusta, Mallotus philippinensis, Cassia fistula, Lagerstroemia parviflora, Litsea polyantha*) and the shrub layer (*Murraya paniculata, Clerodendron infortunatum, Colebrookia oppositifolia, Flemengia semialata, Justicea pubigera*) were completely different (Rana *et al.* 1988) in old growth Sal forest while it showed little variation to the respective layers pertaining to seedling – dominated Sal forest. The presence of different species in different Sal forest represents its type viz. dry, moist or wet and also gives sufficient information about the various edaphic and biotic

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features of the area through their distribution density in the forest (Gautam and Devoe, 2006). Species like *Butea monosperma* and *Mangifera indica* show leaf fall during the months of January to April while that in *Shorea robusta* and *Terminalia tomentosa* lies between March – May. *Syzygium cumini* shows leaf fall in the months of February and March while Cedrela toona experiences the same from November to February. Occurrence of new leaves in *Butea monosperma*, and *Syzygiumcumini* initiates and terminates at same time of the year i.e.from the month of March to May. Leaf formation in Cedrela toona is of only two months (March - April) while in case of *Shorea robusta* and *Mangifera indica* it is from April to July and from July to September in case of *Terminalia tomentosa*. *Syzygium cumini* shows new leaf formation and flowering at the same time (March – May). Flowering season of *Butea monosperma* is the longest (Jan - April) followed by *Mangifera indica (*Feb - April), *Shorearobusta* (March – May), *Terminalia tomentosa* (July - September) and lastly *Cedrela toona* (March - April). Fruiting in all species more or less commences from April (*Butea monosperma, Cedrela toona*), May (*Shorearobusta, Syzygiumcumini)* and June (*Mangifera indica*). Maximum leaf fall occurs during the period of mid February - mid May (Pokhriyal et al. 1987, Singh et al. 1993) followed by seeding which normally ends by mid – June.

Intrinsic factors
Natural Regeneration Problem
Sal regeneration occurs either by seed origination, coppicing or sprouting from root suckers. Equivalent vigour is observed in seedlings of both coppice and seed origin (Troup, 1986). Size of tree affects the size of seed but there is no effect on the viability of the seed. The problem of regeneration in Sal forests has been widely discussed but insufficient and inefficient efforts have been made in this regard till date. Natural regeneration problem shows variability depending upon the pattern of distribution of Sal forests and its types. Poor regeneration potential has been reported in the study conducted by Garkoti et al. 2003, however contrasting results were observed by Tiwari et al. 2010 and Chaubey and Sharma 2013. Regeneration potential may show significant variability in varied environmental conditions. It may be incongruent in protected and unprotected forest regions, natural and managed forests, adjoining areas and geographically separate regions (Deshpande 2015, Awasthi et al. 2015). The key factor for successful natural regeneration of Sal is the profuse quantity of seed supply along with in time commencement of monsoons. Seed supply has been reported to be sufficient in a good seed year while the problem lies in its recruitment and establishment phase (Tiwari 1995). A monograph published by Tewari, 1995 clearly suggests that any disturbance in the site upon commencement of seed fall and rainfall should be avoided as this coincidence period is short and very crucial for the success of regeneration and establishment. In normal natural conditions, *Shorea robusta* seeds are reported to be short lived and upon attainment of maturity the seeds remain viable for the next ten days only and afterwards lose their viability to regenerate (Mishra et al. 2020). The poor recruitment and establishment of Sal saplings varies in different geographical regions. These factors can be summarized as shown in table no.1.

Climate has major influence on the distribution, ecology and the constitution of forests (Agarwal et al. 2018).In addition to the above mentioned features, hydro – meteorological disturbances also have their bearing on the success rate of seedling regeneration. Four distinct monsoonal phases are experienced in India namely southwest summer monsoon (June - August), northeast winter monsoon (December – February), spring (March – May) and autumn (September – November). This spatial variation in monsoon activity is the major causative reason for such a diverse floral distribution in the country. The 1200mm – 1500mmrainfall is considered to be optimal for successful seedling regeneration. Deviance in this range causes abrupt reduction in regeneration potential of Sal forest cover. This trend as reported by Garkoti et al. (2003) in the Shiwalik region and the foothills of Himalayas which showed a decrease in regeneration potential in post monsoon dry periods. A similar trend was observed in the Gangetic plains and Central India with long dry post monsoon periods. The rainfall pattern shows heavy drop off in between monsoon and post monsoon phase. The average annual rainfall also revealed steep descend in the post monsoon season (Mishra et al. 2020). This is important in lieu that post monsoon stage is the period of establishment of *Shorea robusta* seedlings and lower rainfall may have an adverse effect on Sal seedlings. However areas where post monsoon moisture level is retained are ideal for successful regeneration. This shows that dry conditions decrease the regeneration potential at

the establishment stage. On the contrary, hilly areas with appropriate light conditions, proper soil drain and adequate moisture levels, shows profuse natural regeneration in these areas. However in these areas other factors come into play in reducing the regeneration potential. Canopy density and structure management in a proper manner facilitates good seedling establishment and has a direct bearing on the capacity of soil to retain moisture and curb the growth of weeds.

Die back phenomenon

The root system of Sal keeps generating new shoots every year until the development of a root stock strong enough to produce a proper shoot system which eventually develops into a tree. Die back phenomenon is generally associated with the recruitment phase and refers to a disease symptom which is very common in woody plants. It is characterized by the successive decay of branches and shoots from the top. Generally, all trees are susceptible to this phenomenon, in which the premature tree dies or the canopy descends to a poor health condition very rapidly. The major cause of this condition has been observed to be the response against any kind of negative stress exerted on the plant by nematode attack, root borers or heartwood borers infestation to the tree, invasion of defoliators, stem cankers or diseases like thread blight, horse – hair blight, root and collar rot (Tiwari; 1995,Rahman et al. 2010). However, this phenomenon is apparent for a short span of time and the seedlings recover from this situation year after year and grow upon recovery. Thus even under suitable recruitment of new seedlings, if the growth system is irregular as apparent in the die back phenomenon, it may take 30 – 60 years for the proper development and establishment of the next generation. The exact causative factors and their degree of damage to initiate die back of seedlings is not known. The frost injury, caused due to low temperature and heat injury, caused in high temperature regions may lead to die back phenomenon. Also, the seedlings are more successfully established in light areas as compared to shady parts of the forest cover as an appropriate amount of light lowers the risk of die back (Sahu et al. 2005, Mishra et al. 2020) Area specific studies are required to properly understand this phenomenon in different climate provenance.

Seed Factors

Initiation of embryonic growth from a recessive state of dormancy to an active growth life refers to as germination which starts only upon fulfillment of desirable environmental conditions along with the required threshold of light quality (Mishra et al. 2021). Seed germination is temperature and substratum dependent (Anon; 1993). Maximum germination of *Shorea robusta* seed is reportedly attained at 27^0C while minimum germination at40^0C to50^0C. The seed germination at low temperatures may be attributed to secretion of endogamous inhibitors leading to the arrest of embryonic growth or due to imbalance in some enzyme activities due to low temperatures ultimately hindering the formation of nutrition required for embryonic growth (Agarwal et al. 2018).

Extrinsic Factors
Edaphic Factors

Soil moisture, at the seedling stage in particular, along with other factors like soil nutrient availability, aeration and soil erosion have been widely studied for their impact on the natural regeneration process of Sal forest (Gautam et al. 2014). Under the canopy of Sal forest, the vegetation is very scarce and is majorly dominated by shrubs (Timilsina et al. 2007). The prime species present are *Flemingia strobilifera* (L.) *Indigofera pulchella* and *Clerodendrum viscosum* which represent fertile soil composition suitable for regeneration. The soil types in the tropical and sub – tropical forests of the foothills of Central Himalayas particularly are characterized by the alluvial soils and are of two types namely Bhabar and Terai. Bhabar regions contain high water table levels and descend towards the plains, while Terai regions represent alluvial plains followed by Bhabar land (Mishra et al. 2000). Bhabar soil, being well rich is suitable for recruitment of Sal, however, shows difficulty at the establishment stage of seedlings. On the other hand, both recruitment as well as establishment is problematic in the Terai region (Tiwari 1995). Qureshi et al. 1968 suggested in his study that desired light conditions should be properly coordinated with the weeding practices so as to attain better results pertaining to growth and seedling establishment.

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Study conducted by Bhatnagar (1959) revealed the consequences of root competition during the process of natural regeneration and recommended trenching and weeding to be carried out before the peak of seedling growth. Other limiting factors have been realized to be poor aeration, monsoonal dispersion of soil, high levels of magnesium, soil hardiness during dry periods and inappropriate levels of soil moisture etc. (Sharma et al. 1985). Deshpande (2015) reported a negative correlation between physio – chemical properties of soil and soil pH and observed acidic soils to be best suited for healthy Sal Forest regeneration. All these studies focus on an optimal balance of edaphic factors to be available at the root zone of the soil for assuring a healthy regeneration of Sal forests. Apart from that, many studies also have reported the role of microclimate on the regeneration process ((Rautianian and Suoheimo 1997, Sapkota et al. 2009c, Deshpande 2015, Mishra and Garkoti 2015). These studies discussed the features responsible for natural regeneration variables like low temperature, frost, drought, light intensity, soil temperature in winters and so on. Tyagi et al. 2011, studied the effect of variable light intensity and observed that light intensity and the span of illumination available is directly concerned with the resumption process. Tree canopy plays an important part in determining light intensity requirements for regeneration process (Adhikari et al 2017). A study conducted in Nepal showed areas with greater penetration of light radiations at the ground level show better regeneration as compared to areas with dense canopy where light barely reaches the ground (Sapkota et al. 2009, Awasthi et al. 2015).

Shorea robusta is a light demanding tree species. Other than dry conditions, a direct overhead light received from the early stages of establishment ensures the optimal growth. However, shade is also a prerequisite for moisture retention and as a defense against frost (Tiwari 1995). Sal forest cover is associated with different species of trees, shrubs and herbs (Chauhan et al. 2001). These layers of shrubs and herbs are most reliable indicators of forest health and regeneration potential of the climax species of the forest as there exist a substantial competition between the seedlings of *Shorea robusta* and these shrubs and further with other large trees of the forest (Seth and Bhatnagar 1960). Certain communities of Sal show good regeneration potential namely *Sal-Terminalia-Moghania*, *Sal-Lagerstroemia-Pogostemon* and *Sal-Syzygium Randia-Ageratum*, whereas *Sal Ougeinia-Colebrookea* show lower regeneration potential (Bhatnagar 1960). These species compete with other vegetation for growth and utilization of nutrients. *Moghaniachappar* (Benth) and *Murrayakoenigii* (L.) species along with Sal is a good growth indicator whereas *C. viscosum* and *Syzygium cumini (L.)* are growth retarding species which suppress the growth of Sal and hamper nutrient utilization (Srivastava 1972). However both negative as well as positive impact of an associated species may be area specific or may vary according to climatic conditions. For instance, in one study conducted in Doon valley (Pande 1999) reported *C. viscosum;* and other fast-growing species like *S. cumini, Mallotus philippensis* (Lam.) and *Macaranga auriculata* (Merr.) Airy Shaw as negative growth indicators, due to high moisture requirement. On the contrary, in another study conducted by Gautam et al. in 2007, M.*philippensis*, *M. koenigii* and *C. viscosum* proved to be having strong and positive impact on Sal regeneration and negative effect was observed *Adhatodavasica* and *Pogostemon plectranthoides*. However, comprehensive studies in this regard are mandatory to properly understand the interaction nexus of Sal with associated species.

Biotic Factors

Increased incidences of biotic troubles also have their part in altering regeneration potential (Malmstrom and Raffa 2000). Roychoudhary (2015), and around 346 insects and pests species to be associated with Sal, out of which about 155 species are living tree muggers and majority of them belong to the category of defoliators. Sal borer (*H. spini – cornis*) species are oligophagous which feeds primarily on Sal heartwood (Roychoudhary, 2018) and causes severe damage to the standing trees ultimately and silently killing the tree. Its invasion on the tree is characterized by the presence of saw dust near the tree base (Joshi et al. 2002). Trees belonging to all ages with girth more than 20 centimeters are affected by this borer (Bhandari and Singh 1988) with preferable girth range between 91 – 150 centimeters. Thus, these bores directly distress the timber industry (Roychoudhary et al. 2004). It also causes withering out of the tree top branches (Utkarsh, 1998) and needs to be critically managed for sustained supply of Sal timber. A periodic surveillance of borer attack regions has been suggested by Roychoudhary et al. (2017) along with other management techniques like trap tree and removal of infested individuals (Bhandari and Rawat, 2001). Pandey and his coworkers (2020) recommended development and use of chemical formulations to be used as traps to attract

Sal borer beetles which feed on Sal borer and then can be collected thereafter. New techniques and formulations can help control the insect – pest infestations and lead to effective approaches in forest management strategies to be applied against biotic organisms hampering the Sal forest cover and yield. Another biotic factor includes fungal infections like leaf spots and blights namely *Cylindrocladium floridanum* (*aka Calonectria floridanum*) and *C. scoparium*. These have a direct effect on foliage growth of *Shorearobusta* in India (Mehrotra 2001). Major impact is seen in Chhattisgarh and Madhya Pradesh states where root rot disease is more evident in dry and wet Sal forests. This root rot disease is caused by *Inonotus shoreae* (Wakef). Another species reported is *Ryvarden* (previously called as *Polyporus shoreae),* which causes rot on tree tops leading to die back of trees (Jamaluddin, 1991). Trees infected by these fungus, have decaying effect on roots followed by the bark and the sapwood, although the heartwood is unaffected from the ill effects of the fungal diseases. Thus an integrated insect – pest and disease control strategy should be critically appreciated in every conservation program.

Anthropogenic Factors

Human activities have been the foremost cause of habitat fragmentation which alters the forest ecosystem by disturbing the species composition, density and stand structure (McKinney 2002, Deshpande 2015). Overexploitation in Bangladesh has depleted the densely populated Sal forest cover into areas with strewn trees (Rahmann et al. 2010). Haque, 2007 reported a steep decline in the Sal forest cover from more than 60% in late 1970's to nearly 36% in 1985 and thereafter it was reported to be only 10% left in 1990's. Similar trend was seen in other countries like Bhutan, India and Nepal (Acharya et al. 2011, Islam et al. 2012). Illegal felling is another major outcome of demographic load. Gain (2005) reported illegal felling of Sal forest to be as high as 12% in Madhupur region of Bangladesh during the 1990's. For mining purposes, Sal forests have been compromised in Jharkhand state since last few decades (Singh, 2018). Other activities like poaching, growing livestock, encroachment etc. have further worsened the situation for Sal forest cover. Grazing activities lead to hardening of the upper layer of soil and hinder in the emergence process of the seedlings from beneath the soil. These practices are likely to claim the very existence of Sal forest areas, thus needs to be strictly checked and regulated.   Forest fires, frequently used to clear surface weeds and unwanted seedlings, create dry conditions and are not recommended in Sal forests in low rainfall zones (Gautam and Davoe, 2006). Although the positive effect of this practice is the aggravation of new regeneration but it adversely affects the existing saplings. Also it causes loss of top soil nutrients in tropical forests. Frequent forest fires near road sides and human habilitation again worsens the situation manifolds (Adhikari et al. 2017). Forest fires lower the surface flora leading to increased wildlife movement in the area, thus fire incidences need to be properly managed through controlled forest fire strategies (Malla et al. 2018).

Management Strategies

As inferred from the above discussion, it can be clearly observed that natural regeneration can be addressed by the use of critically developed management techniques or good silvicultural methods. Pertaining to silvicultural management related to natural forests, Nepal implemented timber production forestry practices resulting in adequate natural regeneration of Sal (Sah, 2000). Development of community forests also resulted in satisfactory regeneration (Pandyal, 2012). In Indian context, each state Forest Department has developed working plans for the management and conservation of forests.

Artificial Regeneration

These methods include nursery techniques, macropropagation (vegetative propagation) and micro – propagation (tissue culture). *Shorea robusta* seeds are true recalcitrant seed. They tend to lose viability under dehydrated conditions below 37% moisture level (Parkhey et al. 2012). Thus moisture and storage temperature are directly proportional to seed viability of Sal during storage. The viability of seeds can be increased by the use of storage temperature range between 13.5^0C to 23.5^0C (Purohit et al. 1982). Below or above this temperature range, the seeds lose their viability (Tompsett 1985, Khare et al. 1987). This may be attributed to rapid seed moisture loss at hugh (33 - 36^0C) as well as low (5^0C) temperature conditions (Purohit et al. 1982).in addition to this, seed germination and survival of seedling is an attribute of seed size, age of parent tree and positioning of seed into the soil (Patnaik et al.

2015). A recent regeneration protocol involved community participation in sowing the Sal seeds artificially, followed by layering of the pit with termite mound and covering the same with shed Sal leaves in order to retain soil moisture. This practice recorded high level of germination percentage nearly equivalent to 84 - 96% with elevated seed viability up to fourteen days (Agrawal et al. 2018). Another method includes vegetative propagation which overcomes the difficulties arising from recalcitrant nature of Sal seeds (Gbadamosi and Oni 2005). Tiwari (1995), advocated artificial methods like direct sowing, poly pot planting, basket planting, pre – sprouted stump planting, container planting and air – layering. Yet another technique of artificial regeneration is micro propagation method. The advantage of this technique over conventional methods is development of large copies of parent plant material within less time and space. Mass production using nodal explants of *Shorea robusta* have been developed (Singh et al. 2014) and successfully applied (Kumar et al. 2018). These artificial methods give good results but require proper use of advance seed science techniques under critical supervision of silvicultural research programs. Singh and Singh (2019) have done experimentation on establishment of Sal saplings outside its natural habitat and reported better auxin induced better physiological adaptability.

Development of community forests also revealed satisfactory results (Pandyal, 2012). In Indian context, each state forest department has developed working plans for the management and conservation of Sal forests. Various authors have suggested different management practices from time to time. Hole (1921) suggested standardization of game proof fence, maintenance of reasonable equilibrium between shrubs and grasses while, retaining the mid – storey species in order to deal with improper soil working conditions in Sal forests. Trenching and weeding was advocated by Bhatnagar (1959) to deal with root competition problems, while in case of high pH soils and irregular micronutrient distribution, maintenance of optimum soil pH i.e. 4.5 – 5.5 along with practices used to increase the N and K levels and to lower P level in the soil has been advised (Bhatnagar, 1965).Yadav (1966) emphasized on maintenance of proper drainage structure, amelioration, thinning and weeding to overcome the problem of regeneration arising from coarse texture and excessive boulder in soil causing moisture paucity or due to clayey soil resulting in high moisture retention. Adequate light can be received by thinning of canopy (Seth 1967) while the problem related to associated species and nutrition competition can be handled by the use of fertilizers with rich nitrogen content and removal of excessive individuals of associated species (Srivastava 1972). Short seed viability duration can be taken care by coordinated seed fall and rainfall and also through vegetative propagation (Singh et al. 1987).

CONCLUSION

Development of mixed forests with broken canopy helps to promote Sal forest regeneration. Silvicultural practices like thinning at regular intervals of time facilitate in diminishing the excessive canopy and provide proper photoperiods required for *Shorea robusta*. Although, the management techniques for better regeneration of Sal forests, being applied in various parts of the India show limited application and are area specific or problem specific in its construction. Also these methods are somewhat inefficient in the contemporary climatic scenario which has undergone gradual shift from the time when these methods were developed. Earlier SWOT (Strengths, Weaknesses, Opportunities and Threats Analysis) analysis was recognized as an important tool by FAO for forest related research to be applied to gathered, synthesize and analyze the forest data (Kazana et al. 2015). This method has been applied successfully and reported by various authors (Kurttila et al. 2000, Piggin 2003, Harrison and Herbohn 2004, Suh and Emtage 2004, Wong 2005) in different parts of the world. ANR plots, otherwise helpful in protecting the area against wild animals, are within the access of domestic livestock and hence, compromise the management system efficiency. Recent advances in Remote Sensing (RS) and Geographical Information System (GIS) technologies have significantly contributed along with other species modeling tools in the clear and accurate depiction of Sal forest distribution on the map. Global Positioning System (GPS) in combination with a model based technology of Maximum Entropy Distribution (Maxent) is widely used in the approximation of the distribution pattern (Bhandari et al. 2020) which can prove to be of great help in predicting and exploring the possibilities of this species in particular. Numerous factors are responsible for regeneration of *Shorea robusta* having a very fragile equilibrium among them, as discussed

in detail and need to be taken care of for better development and growth of Sal forest cover. New and efficient silvicultural techniques are in demand and are required to be developed with a focus on population genetics, tree improvement, phenology, ecological shifts and other constrains through continuous research, particularly for this magnificent species.

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Table No 1: Summary of various factors affecting natural regeneration

S.No	Factors	Types
1	Climatic factors	Humidity, temperature, light intensity, span of light receiving hours, precipitation, rainfall and wind.
2	Edaphic factors	Depth, aeration, moisture level, nutrients and erosion.
3	Seed factors	Seed sensitivity, poor seed viability and seed dispersal.
4	Biotic factors	Wildlife activities, insect and pest infestations, disease and grazing activities by animals.
5	Abiotic factors	Forest fires, over browsing and water deficiency
6	Anthropogenic factors	Illegal felling, encroachment, deforestation, poaching

Table No 2: Common species which attack *Shorea robusta*

S.No.	Type	Species
1	Root Borers	*Pammene theristhis*
2	Heartwood Borers	*Hoplocerambyx spinicornis*
3	Defoliators	*Lymantria mathura and Ascotisimperata*
4	Stem Canker	*Macrophoma shoreae*
5	Thread Blight	*Polyporus sp.*
6	Horse-Hair Blight	*Marasmius gordipes*
7	Root And Collar Rot	*Xylaria tuberosa and X. polymorpha*

RESEARCH ARTICLE

Enhanced Analysing Node Behaviour

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ABSTRACT

One of the mobile nodes should carry out the unpredictable packet sharing to its next neighboring node since the energy level of a node is changed. Consequently, those nodes need to increase its communication rate else minimize its communication rate from source node to a destination node, the multipath routing is also not easy to provide the lesser rate of packet transmission of a specific node. If energy level not sufficient the protocol selects an alternate path to carry on routing using analyze the node behavior successfully. The proposed approach takes primary parameters of a node, and each parameter is described with its equation to reach the efficiency in the lifetime of the network. that nodes are close and the link is sufficiently stable. Otherwise, it calculates LS using equation . The downstream node on receiving the route it updates its routing table. It routes the data packets in the new route available, preventing packet losses due to link breakage. Performance analysis of the proposed EN_AOMDV protocol is finished using the NS2 version 2.35; Network lifetime is a time at which the first node in the network is running out of energy to participate in communication process all mobile nodes in the Ad hoc network environment are independent of the environment also which are performed its choice in communication.

Keywords: communication, equation, environment, downstream.

INTRODUCTION

The mobile nodes are having a difference to each other available node in a network environment because its nodes characteristics and capability is different. One of the mobile nodes should carry out the unpredictable packet sharing to its next neighboring node since the energy level of a node is changed. Consequently, those nodes need to increase

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its communication rate else minimize its communication rate from source node to a destination node, the multipath routing is also not easy to provide the lesser rate of packet transmission of a specific node. The Node support is measured as an essential unit for preserving system link in an unformed structure such as MANETs because all mobile nodes in the Ad hoc network environment are independent of the environment also which are performed its choice in communication. In order to monitor measuring the crash of breakdown mode in the direction of the link of the network that is demonstrated by the subsequent process. Regard as one node needs to share data packets with the other node in a MANETs, and another node starts to send a request to the next neighbor for sequential manner. Since the condition is a node breakdown, then the neighbor node is not capable of transmitting, therefore carry out the route discovery process to its further next nodes in that path. Therefore, the path does is not recognized among the sender and destination node along with the neighboring nodes. A breakdown node that relays the path finding request packet to the next node, except it, deny to transmitting the data packets to following the link available path..

ENHANCED ANALYSE NODE BEHAVIOUR
This study proposes enhanced analyze node behavior protocol that the Node support is measured as an essential unit for preserving system link in an unformed structure such as MANETs. The proposed protocol aims to improve the analyze the nonlinear optimization method for classifying nodes into the breakdown and normal nodes, which are used sequential chance rate, investigation for identifying the breakdown nodes.

Problem Identification
The analyze node energy level also plays a crucial role in MANETs routing. Focus is on identifying analyzing nodes behaviour level consumed therefore far, and energy level leftover and higher than the threshold value assumed to be half the initial value of the nodes energy assumed, which should be sufficient for performing the upcoming transmission, data communication, node route discovery, route selection at the destination. If energy level not sufficient the protocol selects an alternate path to carry on routing using analyze the node behavior successfully.

- Node Stability Aware Metric
- Node Energy-Aware Metric
- Reliablepathfordatacommunicationbasedonroutestabilityandresidualenergymetrics
- System Link and Node Support & Node Route Discovery
- Route Discovery at Intermediate Nodes
- Node Route selection at Destination
- Route node character Maintenance

The node energy level efficient parameters in this proposed protocol are proposed based on the evaluation of effective individual performance. By combining these node energy level efficient parameters, the objective of the proposed work is reached .

Node Stability Aware Metric (NSAM)
NSAM model considers signal strengths and mobility for computing the probability of link failures. It computes Link Stability (LS) using signal strength values received from the MAC layer.

Node Energy-Aware Metric (NEAM)
It is assumed that all wireless nodes come with the residual power detection device. The source application layer communicates n value to the network layer for selecting nodes that meet the energy requirement. It avoids linking breakages due to energy depletion. The aim of this metric is to maximize NEAM. It takes the product of the residual battery of the intermediate nodes to select a path that has nodes with maximum residual energy among the path that just meet the essential energy requirement REQe .

Reliable path for data communication based on route stability and residual energy metrics

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To find a reliable path for data communication based on route stability and residual energy metrics. The above bi-objective optimisation problem can be transformed into a single objective problem, by providing importance factor (i.e. W1 and W2) for each criterion of the objective.

System Link and Node Support & Node Route Discovery

When a node S needs to send packets to a destination D, it searches for the route in its route table. If a route to the destination D is not available, then the source S will broadcast a Route Request (RREQ) message to its neighbours. The RREQ of Energy Ad-Hoc Multipath Distance Vector (EN-AOMDV) is an extension of an RREQ packet of PAAODV routing protocol .

Route Discovery at Intermediate Nodes

If the strength of the RREQ packet is weak, then it drops the RREQ. Then node i checks whether its residual energy will meet the required energy REQe specified in the RREQ packet.

Node Route selection at Destination

Destination node will receive RREQ packets from different possible routes. On receiving the first RREQ packet, the node D starts a timer t1 for the duration of Route Reply Latency (RRL) time. It stores all the RREQ that arrives, in its routing table. It computes RF value for the path explored by the RREQ.

Route node character Maintenance (RNCM)

In dynamic mobile Ad-hoc networks, link-breaks occur frequently. EN-AOMDV comes with a make-before-break route maintenance mechanism. This mechanism quickly adapts to the link breakage likely to occur due to the mobility and energy drain.

METHODOLOGY

This study proposes an Enhanced Energy Adhoc Mobile Distance Vector (EN_AOMDV) protocol to improvise the functionality of the node behaviour and signal strengths for computing the probability of link failures, residual power detection, route stability and residual energy metrics, system link and node support, route discovery. The proposed analyse the node behaviour process follows the procedures of PAAOMDV protocol with the help of energy-efficient parameters. The proposed approach takes primary parameters of a node, and each parameter is described with its equation to reach the efficiency in the lifetime of the network. The study has chosen the multiple CPUs feature as a primary parameter for the load balancing of the proposed approach. Following are the few assumptions considered for the proposed approach.

Node Stability Aware Metric (NSAM)

NSAM model considers signal strengths and mobility for computing the probability of link failures. It computes Link Stability (LS) using signal strength values received from the MAC layer. Any link e has an associated link stability LS(e) and it is given by

$$\text{LS}ij = \frac{u2 - DSSij}{u2 - u1} \qquad \text{(equ.4.1)}$$

Where DSS is the differentiated signal strength to decide whether the signals are getting stronger or weaker. It is computed as follows.

$$DSS\,i,j = SScur\,i,j - SSnew\,i,j \qquad \text{(equ.4.2)}$$

A path between source s and destination d is given as P(s, d) = (s, e(s, x), x, e(x, y),y, e(z, d), d). Formally, a path between two nodes s and d is a set of all feasible path between them and can be represented as P(s, d) = P0, P1, . . .,

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Pn, where each Pi is a feasible path between s and d .
Equation 4.3 define the stability of the path P, by the product of link stability of its edges as follows

$$\text{Stability (p)} = \prod_{e \in p} LS(e) \qquad \text{(equ.4.3)}$$

Node Energy-Aware Metric
It is assumed that all wireless nodes come with the residual power detection device. The energy required to transmit a packet (Etx) can be computed as

$$Etx = \frac{Psize * Ptx}{BW} \qquad \text{(equ.4.4)}$$

where Psize is the packet size, Ptx is the packet transmitting power and BW is the bandwidth of the link. The transmitting energy is directly proportional to the distance between nodes. The source application layer communicates n value to the network layer for selecting nodes that meet the energy requirement. It avoids link breakages due to energy depletion. The Total Energy Required (REQe) for data packet transmission is given by

$$REQ\,e = n * (Etx + Epr\,oc) \qquad \text{(equ.4.5)}$$

$$EM(p) = \prod_{i=1}^{n} Ri / Fi \qquad \text{(equ.4.6)}$$

where Ri(t) is remaining battery capacity and Fi is the full battery capacity of intermediate node i, at time t. The aim of this metric is to maximise EM. It takes the product of the residual battery of the intermediate nodes to select a path that has nodes with maximum residual energy among the path that just meet the basic energy requirement REQe . Reliable path for data communication based on route stability and residual energy metrics To find a reliable path for data communication based on route stability and residual energy metrics. The above bi-objective optimisation problem can be transformed into a single objective problem, by providing importance factor (i.e. W1 and W2) for each criterion of the objective. combine the objectives into a single objective function to calculate the Reliability Factor (RF) of the path P, can be mathematically stated as

$$RF\,(P) = W1 \cdot Stability(P) + W2 \cdot EM(P) \qquad \text{(equ.4.7)}$$

where the parameters w1 and w2 are chosen based on the network dynamics and application requirements. In this study, to giveequal importance to both stability and energy metrics, it is assigend assign 0.5 to both W1 and W2, such that W1+ W2=1 condition is satisfied(Kandhalu et al, 2010). Consequently, the sum of the objectives must be maximised and Maximum Reliability Factor (MRF) can be computed by

$$MRF = max(RF(P1), RF(P2)..., RF(Pn)) \qquad \text{(equ.4.8)}$$

The path with MRF value is selected as a reliable path for data transmission.

System Link and Node Support & Node Route Discovery
The RREQ of RSEA-AODV is an extension of an RREQ packet of an EN_AOMDV routing protocol. Three new fields Accumulated Path Stability (APS), Accumulated Energy Metric (AEM) and required energy (REQe) are added to the RREQ packet. It initialises the values to the added fields as follows: APS, AEM with 1. e) Route Discovery at Intermediate Nodes If the strength of the RREQ packet is poor, then it drops the RREQ. Then node i checks whether its residual energy will meet the required energy REQe specified in the RREQ packet. If the above conditions are satisfied, then node i make a reverse route entry in the Routing Table (RT). Then it calculates LS. If the signal

strength is above SThr1, then it assigns 1 to LS. It implies. that nodes are close and the link is sufficiently stable. Otherwise, it calculates LS using equation . After these steps, it updates the APS and AEM fields such that the updated values contain the route stability and energy metric of the explored route up to the current node. It enters the relevant information from RREQ into Route Request Forward Table (RFT). Then, node I broadcast the RREQ to its neighbour. In case of the duplicate packet, if it contains better values for APS or AEM, then it makes an entry in RFT and discards the packet. On receiving an RREP packet, node i measures the strength of RREP. If its strength is weak, then it will drop the RREP packet. It looks up RFT for the corresponding RREQ entries, to select the node with the highest RF value. It forwards the RREP packet to the node with the highest RF value. It is shown in Algorithm 2. It makes an entry in the RT

Node Route selection at Destination

Destination node will receive RREQ packets from different possible routes. On receiving the first RREQ packet, the node D starts a timer t1 for the duration of Route Reply Latency (RRL) time. It stores all the RREQ that arrives, in its routing table. It computes RF value for the path explored by the RREQ. If destination nodes receive more than oneRREQ before the timer t1 expires, then it forwards the RREP packet to the node with the highest RF value. It is shown in Algorithm 3. This reduces the amount of control overhead incurred during the route establishment due to multiple RREP for single RREQ as in AOMDV . The Destination node makes an FT entry for the flow. In case, if it does not receive any data packets within the timeout, then it will delete the respective entry from the FT table. Destination node will receive RREQ packets from different possible routes. On receiving the first RREQ packet, the node D starts a timer t1 for the duration of Route Reply Latency (RRL) time. It stores all the RREQ that arrives, in its routing table. It computes RF value for the path explored by the RREQ. If destination nodes receive more than one RREQ before the timer t1 expires, then it forwards the RREP packet to the node with the highest RF value. It is shown in Algorithm 3. This reduces the amount of control overhead incurred during the route establishment due to multiple RREP for single RREQ as in AOMDV . The Destination node makes an FT entry for the flow. In case, if it does not receive any data packets within the timeout, then it will delete the respective entry from the FT table.

Algorithm:1 Implemented in Intermediate Nodes.

Input: A RREQ packet P from the neighbour
node.
Step 1: if (Node Battery <EThru) or (SSnew <SThr2) then
Step 2: Drop Packet P
Step 3: end if
Step 4: if ((RREQnotalready forwarded) or (RREQhasbetter APS or AEM value))
Step 5: then
Step 6: if (SScur>SThr 1) Then LS -1
Step 7: DSS – Sscur – Ssnew
Step 8: if (SSnew<SThr 1) and (SSnew> SThr2) then
Step 9: if (DSS <u1)Step10: then
Step 11: LS – 1
Step 12: else
Step 13: LS – (u2 – DSS)/ (u2 – u1)
Step 14: end if
Step 15: end if
Step 16: PAS - APS * LS
Step 17: AEM – AEM * {{RBCi/FBCi}}
Step 18: Update RREQ with APS and AEM
Step 19: Broadcast RREQ packets to the next hop
Step 20: Else
Step 21: Drop Packet P

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Step 22: end if

Algorithm:2 Implemented in Intermediate nodes
Input: A RREP Packet P from Neighbouring Node
Step 1: Line (1 – 3) of Algorithm 1
Step 2: N = number of the entry in RFT for corresponding RREQ
Step 3: F = Index of the first entry in RFT for corresponding RREQ
Step 4: I = F
Step 5: Count = N
Step 6: While(Count >1) do
Step 7: J = Next entry in RFT for that RREQ
Step 8: if (w1*RFT|j|.APS + W2 * RFT|j|.AEM)>RELFACT)
Step 9: then
Step 10: I = j
Step 11: end if
Step 12: Count = Count – 1
Step 13: end while
Step 14: S= RFT[i].prevhop
Step 15: Make an entry in the RT table
Step 16: Delete the RFT entries for the corresponding RREQ
Step 17: Forward RREP packet to S
Step 18: end if

Algorithm:3 Implemented in Intermediate nodes
Input: A RREQ Packet P from Node N with APS and AEM
Step 1: Line 4 -8 of Algorithm 1
Step 2: N*=N
Step 3: MRE:=W!*APS+W@*AEM
Step 4: Start timer (t1) *for 1st copy of RREQ only*
Step 5: While (! timer – out) do
Step 6: if (a RREQ arrives at D) then
Step 7: NEWRF: w1*APS + w2*AEM
Step 8: if (NEWRF>MRF) then
Step 9: MRF: =NEWRF
Step 10: N*=N
Step 11: end if
Step 12: end if
Step 13: end while
Step 14: Make an entry in RT and FT
Step 15: Send a RREP packet to N*

Route node character Maintenance
In dynamic mobile Ad-hoc networks, link- breaks occur frequently. EN-AOMDV comes with a make-before-break route maintenance mechanism. This mechanism quickly adapts to the link breakage likely to occur due to the mobility and energy drain . It is depicted in Algorithm 4. It executes the mechanism after every t2 seconds, to monitor the status of the route established. If an intermediate node is in the critical battery status or it is receiving weaker signal packets, then it creates an HLP message with Time To Live (TTL) set to 1 and broadcasts it to its neighbours. Neighbours on receiving the HLP packet, check for route availability in its routing table for the destination specified in the HLP packet. If the route is available, then it returns the route to the downstream node of

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the node broadcasting the HLP packet. The downstream node on receiving the route it updates its routing table. It routes the data packets in the new route available, preventing packet losses due to link breakage.

Algorithm: 4 Route Maintenance by Make –before-break mechanism
Input: A packet P from a neighbouring node
Step 1: Executed periodically after timer t2 expire
Step 2: if (SSnew<SThr2) or (Node Battery <Ethr) then
Step 3: if (intermediate node)
Step 4: then
Step 5: Send HLP Packet to all its 1 hope neighbours for an alternate path
Step 6: else//if destination node
Step 7: Send Stop – Traffic intimation to the source node
Step 8: end if
Step 9: end if
Step 10: if (timeout)//Alternate route not found
Step 11: send RCR to the source Node
Step 12: end if

If the node with critical battery is the destination node, then it will send the stop traffic intimation to the source node, to avoid future packet drops and wastage of resources. If there is no alternate route available with the one-hop neighbours, then after the expiry of timer, i.e. timeout, the node will send the Route Change Request (RCR) to the source node. The source node on receiving the RCR will go for the re-route discovery to the destination. In order to identify these unreliable nodes (running low in energy level) using the energy level values calculated for each node. The node energy level value calculation is based on the parameters shown in Table.1.

Node energy calculation is based on nodes sending and receiving rate. If a node is selected for transmission, then it should concentrate more on the corresponding transmission to save energy and not to drain out by involving in unnecessary transmissions. In order to identify energy level, the nodes are evaluated where sender to increase radio frequencies to identify best nodes with more energy levels. The current energy level of a node can be calculated by the initial energy level and the consumed energy level of a node. Setback in energy-based node work is that the source itself may drain out. In such cases introduce external energy to the source node by introducing virtual energy concepts. Other nodes have to store energy for future transmissions .

Analysing The Node Energy Value Calculation Procedure

Step 1: Set initial parameters values as initial energy = 100, maxenergy=0, nodes=50 and Nodeid (unique id for each node)
Step 2: Calculate Intermedenergy based on event, time where events can be (event = "r" || Event= "d" || event ="s"|| event="f")
Step 3: Compute consumed energy for each node For (i in Intermednergy) {
　　　consumenergy[i]=initialenergy-Intermedenergy[i]
　　　totalenergy +=consumenergy[i]
　　　if(maxenergy<consumenergy[i]){
　　　maxenergy=consumenergy[i]
　　　nodeid=i }}
Step 4: Compute average energy Average energy=total energy/nodes.

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Higher Residual Node Energy Based Route Analysing

Stable multicast routes are constructed with the help of neighbor node selection mechanism in which the next node is selected based on residual energy and density. In the following section, RQ and RP packet format, the process of request and reply phase, route establishment process and route maintenance are discussed in Figure.1.

Request packet format details: Request packet format includes of following components

◎ **SRCIP**: It is the address of the source from where the path needs to be established to the multicast destinations.
◎ **MGIP:** Multicast group address where destinations are present as group members.
◎ **Seqno:** Packet identification number set at source node to receive packet sequentially to avoid duplication of request packet at intermediate nodes.
◎ **Hop Count (HC):** Maximum integer value set by the source node and its value decrease by one after visiting each node. It is used to discard the packet when HC reaches zero and inform the same to its source node.
◎ **Time Stamp:** Waiting time of nodes set at source; if next hop was not found, then the node will wait for given timestamp value.
◎ **Visited History:** It stores the address of all previously visited nodes by RQ packet from source to destinations.
◎ **Power Level**: Minimum power level of a node whose remaining power is higher than the threshold value, and this field get an update when the power level is less than the threshold value.
◎ **Node Density:** It is the ratio between node degree and transmission range. It is a positive integer which represents the maximum number of neighbours that a node can support in a multicast routing. It is inversely proportional to the average hop count and directly proportional to power consumption.
◎ **Flag:** Packet identifier. Flag value is set to 1 to recognise RQ packet

Reply packet format details: Reply packet format comprises of following components

◎**SRCIP:** Address of source node from where RQ packet routed.
◎ **DestIP:** Address of destination node where RP packet originated.
◎ **Seqno**: Packet identification number set by the node where RP packet routed, and it helps the source to identify RP for RQ packet sent.
◎ **Hop Count (HP):** Maximum persistent integer value set by destination node and the count is equal to the number of nodes in the visited history field traced by RQ packet.
◎ **Time Stamp:** Set by destination node and node searches next hop to forward RP packet, if not found then node will wait for given time stamp value.
◎ **Visited History:** Visited previous nodes recorded in visiting sequence. It stores a sequence of addresses traced by RQ packet and will be used by RQ packet originator with the help of RP packet.
◎ **Power Level:** Final minimum power level of node brought by RQ packet at the destination and it remains constant until RP packet reaches to the source node.
◎ **Node Density:** Degree of intermediate nodes brought by RQ packet and this value is remaining constant until RP packet reach to source node.
◎ **Flag:** RP Packet identifier. Flag value is set to 0 to recognise RP packet .

Three types of databases maintained at each node in multicast routing, namely, (Table 4.2) temporary history table to store visited history of RP packet at each node. (4.3) Neighbour table to prune inefficient neighbour node. (4.3) Routing table to successfully route the data from the source node to the destination node in a multicast group. The tabular form of the database used in the scheme is as follows.

Request Phase

A source node finds the route to its group of destinations by using RQ packets. The sequence operation that occurs in a route request phase is as follows. In phase (1) Source node prepares a RQ packet with residual energy and node density. In phase (2) Selective transmission of RQ packet to neighbours who satisfy selection criteria (power levels ith), and node density requirement. In phase (3) intermediate receiving node discards RQ packet if it is already received (using sequence number in RQ packet) . In phase (4) if RQ packet is not a duplicate, checks Routing

Information table for the availability of route if available, RP packet will be generated and start reply propagation to source. In phase (5) if RQ packet is a duplicate, then discard it and stop transmission of RQ packet. In phase (6) if not duplicate and no route available in Routing Information table, transmit the RQ packet by updating its fields (route record, computed power level, node density, time stamp, and next-hop address) to its neighbours as in step 2. (7) Perform steps 3 to 6 until the destination is reached, and in phase (8) if the receiver is not reached within specific hops, sends RQ packet back to the source node . d) Reply Phase: Multicast destinations initiate the reply phase. When RQ packet reaches the destination node, the following sequence of operations are taking place in the reply phase. In phase (1) RP packet is generated from RQ packet by performing following changes in RQ packet; destination and source node addresses are interchanged, route record is reversed, updates visited history records residual energy record as power level, node density record as node density. In phase (2) update route information at the destination node with destination IP, path information, power level, node density and time. In phase (3) RP packet is forwarded to the next-hop node as per the route record route information table if power level and node density are satisfied. (4) The source that receives RP packet checks whether available power level is more significant than the threshold value, and node density is less than the threshold if therefore, updates routing table by using contents of RP packet .

Updates will only happen if the current time is greater than the time recorded in the routing table. If remaining energy is less then threshold value and node density is greater than the threshold, sends RQ packet to the destination and visited intermediate nodes to stop RP packet propagation. In phase (5) perform steps 3 and 4 until the source is reached. In phase (6) if the source is not found due to link breaks, send RQ packet to the destination, and in-phase (7) the source node chooses one of the received paths with higher residual energy and less node density and keep remaining paths as backup paths. Figure.1 illustrates the essential operation of request and reply phase from source S to destination D3, through the set of intermediate nodes I1.

Route Maintenance

Route maintenance is required in case of link or node failures. Failure of node and link is significantly less because the scheme constructs the stable path from source to destination maximise full connectivity in the network using a larger lifetime of the nodes. However, rarely three types of link failures: link failure between stable intermediate nodes, link failure between source and stable intermediate node, and link failure between destinations and stable intermediate node. In that case, the network problem could be in the following ways. (1) In case of link failure between two stable intermediate nodes, the node detecting failure condition will use RQ and RP packets to find the stable path between itself and the destination. The new path from the intermediate node to the destination will be informed to the source where RQ packet originated in Figure.1 . If a new path is not found, the node sends RQ packet to the source to rediscover the paths. (2) In case of link failure between source and stable intermediate node, the source node will probe the backup path, if it is working, it will use backup path. Routes will be rediscovered if backup path does not exist. (3) In case of link failure between destination and stable intermediate node, the intermediate node will use RQ and RP packets to discover paths to the destination from itself and informs the source about the path. If the route is not discovered, the node sends RQ packet to the source to initiate route rediscovery. The source constructs a new path in all the cases for further routing of packets

RESULTS AND DISCUSSION

Performance analysis of the proposed EN_AOMDV protocol is finished using the NS2 version 2.35; Network lifetime is a time at which the first node in the network is running out of energy to participate in communication process all mobile nodes in the Ad hoc network environment are independent of the environment also which are performed its choice in communication. Monitor the power consumption, Node Stability, Node Energy, System Link, route discovery, Node Route selection of individual nodes; therefore, that nodes will not die due to energy exhaustion. For the given problem statement, to monitor the power consumption, Node Stability, Node Energy, System Link, Route Discovery, Node Route selection of individual nodes and measuring the crash of breakdown

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mode in the direction of the link of the network. This study motivation is that one node needs to share data packets with the other node in a MANETs. Then another node starts to send a request to the next neighbour for sequential manner. In the simulation, Since the condition is a node breakdown, then the neighbour node is not capable of transmitting, therefore carry out the route discovery process to its further next nodes in that path. This study has also compared the obtained results with the PAAOMDV protocol. Figure 2 and 3 presents the comparison results of packets received among the proposed protocol EN_AOMDV, and the existing. AOMDV_LP with 100 and 200 nodes. EN_AOMDV consumes 39% of packets received for 400 rounds. Whereas, the existing protocols AOMDV_LP and MMRE_AOMDV consume 62%, PAAOMDV 63% and 61% respectively.

In conclusion, this proposed protocol EN_AOMDV improves 11% in 100 nodes and 15% in 200 nodes of packets received than the existing AOMDV_LP, which consumes the minimum no of packets from end to end of the existing three protocols. Figure 4 and 5 presents the comparison results of the total number of packet loss among EN_AOMDV and the existing protocols AOMDV_LP, MMRE_AOMDV and PAAOMDV after the completion of 400 rounds. The total no of packet loss of proposed EN_AOMDV protocol and the existing protocol AOMDV_LP, MMRE_AOMDV and PAAOMDV are taken for 100 and 200 nodes. In conclusion, this proposed protocol EN_AOMDV packet loss 10% in 100 nodes and 13% in 200 nodes and of lesser messages than the existing PAAOMDV protocol, which receives the highest no packet loss of the three existing protocols.

Figure 6 and 7 presents the comparison results of the total number of packets delivered among EN_AOMDV and the existing protocols AOMDV_LP, MMRE_AOMDV and PAAOMDV after the completion of 400 rounds. EN_AOMDV receives 11820 messages. Whereas, AOMDV_LP receives 8970 messages, MMRE_AOMDV 10850 messages and PAAOMDV 10781 messages. The total no of received messages of proposed EN_AOMDV Protocol and the existing protocol AOMDV_LP, MMRE_AOMDV and PAAOMDV are taken for 100 and 200 nodes. In conclusion, this proposed protocol EN_AOMDV receives 48% in 100 nodes and 27% in 200 nodes of higher messages than the existing AOMDV_LP protocol, which receives the highest no messages of the three existing protocols.

Figure 8 and 9 presents the comparison results of Residual Energy among EN_AOMDV and the existing protocols AOMDV_LP, MMRE_AOMDV and PAAOMDV after the completion of 400 rounds. The total no of residual energy of proposed EN_AOMDV Protocol and the existing protocol AOMDV_LP, MMRE_AOMDV and AAOMDV are taken for 100 and 200 nodes. In conclusion, this proposed protocol EN_AOMDV receives 21% in 100 nodes and 10% in 200 nodes of energy higher than the existing AOMDV_LP protocol, which receives the highest no energy of the three existing protocols.

CONCLUSION

This chapter has discussed the protocol is applied to monitor on measuring the crash of breakdown mode in the direction of the link of the network that is demonstrated by the subsequent process. Analyse the nonlinear optimization method for classifying nodes into the breakdown and normal nodes, which are used sequential chance rate, investigation for identifying the breakdown nodes.

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Table 1. Analyse of Node Energy Value Calculation Parameters

Parameters	Description
Network Space	100 X 100
Number of Nodes	100,200
Initial Energy	1 Joule (J)
Packet Length	500 bits
Physical layer	IEEE 802.11
Node speed	varied [0m/s - 10m/s]
Pause time	5s
Hello interval	2.0s
TC interval	5s
Transmit power	0.001w
Simulation Time	400 Seconds
Mobility Model	Random Way Point
Protocol	AOMDV

Table 2: Temporary Node Visited History Table

MG Address	Sqno	Hop Count	Path Info
128.16.10.1	4	5	129.16.100.1

Table 3: Neighbour Node Information

Neighbour Address	Power level	Node density	PNF
128.16.10.1	4	5	129.16.100.1

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Table.4: Routing Information Table

MG Addr	Dest.addr	next	hop power level	pi(t)
128.16.10.11	128.16.10.1	128.16.100. 10	40	0.01

Table 5. End-to-End Delay for 100 and 200 Nodes

Time (Seconds)	Existing (MMRE_OMDV)	Existing (LN_AOMDV)	Existing (PAAOMDV)	Proposed (EN_AOMDV)
End-to-End Delay for 100 Nodes				
10	1.04	1.08	1.12	0.82
25	1.08	1.11	1.19	0.84
50	1.14	1.18	1.23	0.89
75	1.42	1.48	1.52	0.96
100	1.53	1.59	1.61	0.98
End-to-End Delay for 200 Nodes				
10	1.06	1.09	1.14	0.84
25	1.10	1.12	1.20	0.85
50	1.16	1.20	1.25	0.91
75	1.44	1.49	1.54	0.98
100	1.55	1.62	1.64	0.99

Table 6. Packet Lost for 100 and 200 Nodes

Time (Seconds)	Existing (MMRE_OMDV)	Existing (LN_AOMDV)	Existing (PAAOMDV)	Proposed (EN_AOMDV)
Packet Lost for 100 Nodes				
10	16	13	10	9
25	63	52	46	26
50	92	85	77	62
75	143	132	121	86
100	151	39	148	102
Packet Lost for 200 Nodes				
10	17	15	12	10
25	62	54	56	28
50	91	82	78	60
75	137	129	118	87
100	151	39	148	102

Table 7. Packet Delivery Ratio for 100 and 200 Nodes

Time (Seconds)	Existing (MMRE_OMDV)	Existing (LN_AOMDV)	Existing (PAAOMDV)	Proposed (EN_AOMDV)
Packet Delivery Ratio for 100 Nodes				
10	25	27	29	34
25	39	41	48	59
50	70	73	79	82
75	83	87	90	91

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100	90	93	95	97
Packet Delivery Ratio for 200 Nodes				
10	26	25	30	36
25	40	43	49	61
50	72	75	81	85
75	85	88	92	93
100	91	94	96	99

Table 8. Energy Consumption for 100 and 200 Nodes

Time (Seconds)	Existing (MMRE_AOMDV)	Existing (LN_AOMDV)	Existing (PAAOMDV)	Proposed (EN_AOMDV)
Energy Consumption for 100 Nodes				
10	27	19	15	8
25	39	32	25	21
50	55	49	44	36
75	72	74	67	48
100	92	85	72	53
Energy Consumption for 200 Nodes				
10	29	21	17	9
25	37	31	24	20
50	53	47	42	33
75	71	72	64	44
100	90	83	70	52
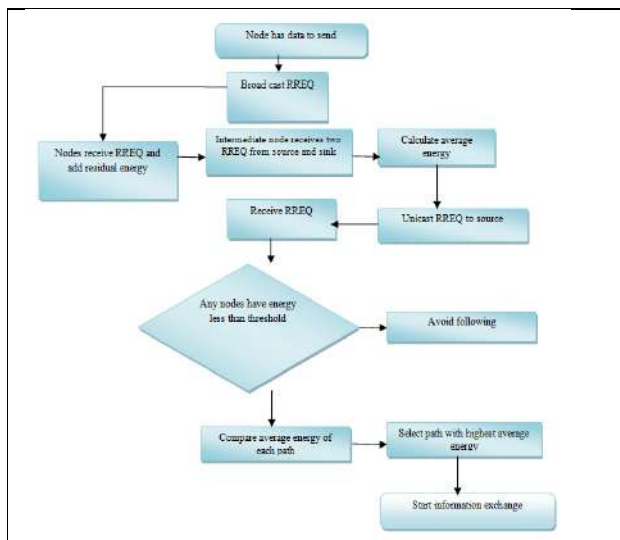
Figure 1. Process Flow in Proposed Analyse Node Behaviour	**Figure 2. Total Numbers of Packets Received for EN_AOMDV Protocol for 100 nodes**
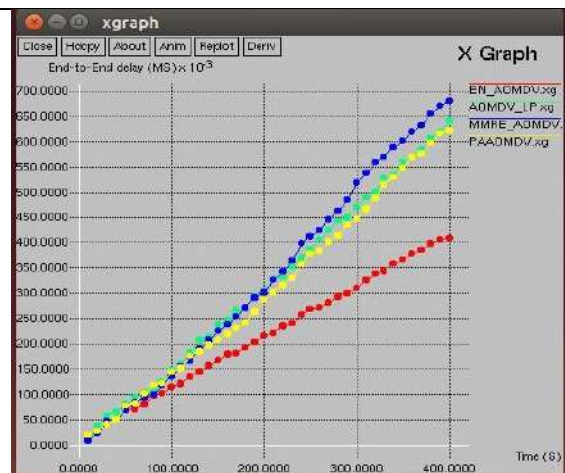
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Figure 3. Total Numbers of Packets Received for EN_AOMDV Protocol for 200 Nodes
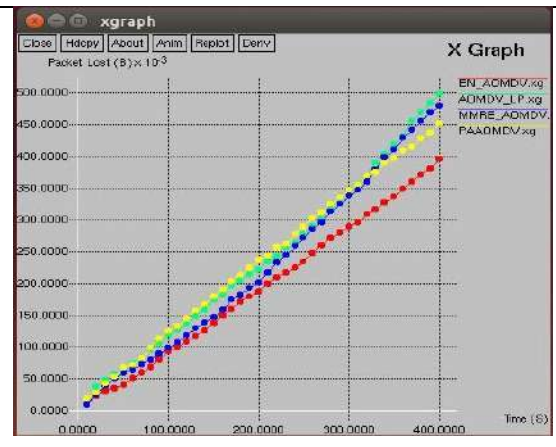
Figure 4. Total Numbers of Packets Loss for EN_AOMDV Protocol for 100 nodes

Figure 5. Total Numbers of Packets Loss for EN_AOMDV Protocol for 200 Nodes
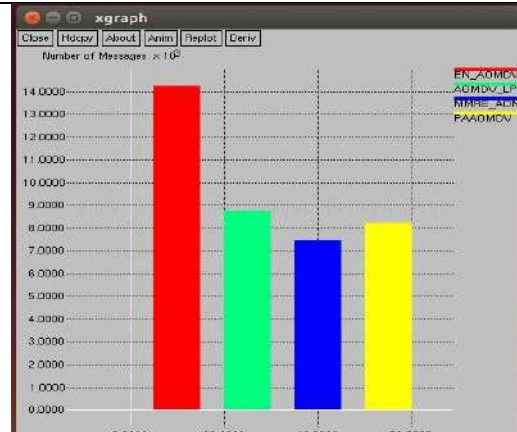
Figure 6. Total Number of Packet Delivered for EN_AOMDV Protocol for 100 nodes
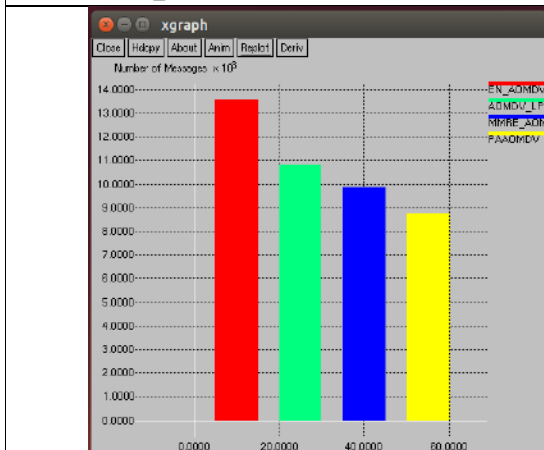
Figure 7. Total Number of Packet Delivered for EN_AOMDV Protocol for 200 nodes
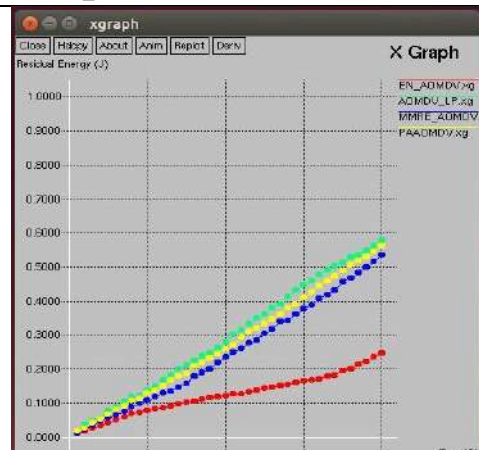
Figure 8. Residual Energy for EN_AOMDV Protocol for 100 nodes

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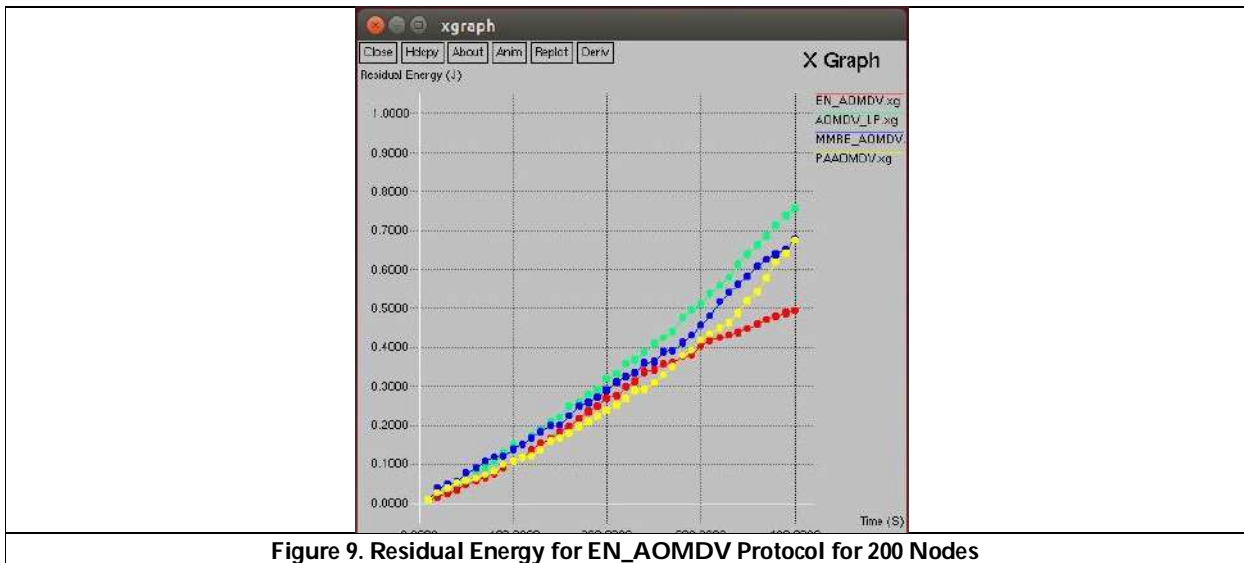
Figure 9. Residual Energy for EN_AOMDV Protocol for 200 Nodes

RESEARCH ARTICLE

Regulatory Aspects of Fragrance in Cosmetics

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ABSTRACT

Fragrances are an intricate blend of natural and synthetic compounds that are applied to products to impart a distinct odour. It can be present in a large number of consumer products including cosmetics, personal care, and household washing products, in addition to perfumes. They meet important emotional demands while also addressing practical issues like hiding unpleasant odours. Consumer companies frequently depend on fragrance development to distinguish their products and provide benefits, and smell can be a crucial driver of consumer choice. Various fragrance components, as well as other substances used in fragrance compositions, have been demonstrated to have negative effects. Some of them are prohibited or restricted in cosmetics under the country's regulations. Other international organizations that focus on fragrance chemical safety, for example, the International Fragrance Association and the Research Institute for Fragrance Materials, publish studies on whether or not particular fragrance chemicals are suitable for use in cosmetics based on their toxicity. This review article aimed to highlight the regulatory aspect of fragrances in cosmetics in the US and EU. The article also focussed on Ingredient safety and labelling requirements for fragrance chemicals in cosmetic products.

Keywords: Fragrance ingredients, Cosmetics, Perfume, Regulations, USFDA, EU

INTRODUCTION

Fragrances are intricate blends of natural and/or synthetic compounds that are applied to consumer products to impart a distinct odour. It can be found in a large number of consumer products to give them a pleasant odour, cover

the inherent odour of some other substances and increase the overall experience of the products[1,2]. Fragrances are a mix of science and art in which chemists become creators and molecules become memory makers. They're created with thought, passion, and ingenuity. They bring joy to billions of people all over the world who use and enjoy fragrance daily, from a fresh-smelling shampoo to a fashionable perfume, from a scented candle to newly laundered linens [3]. Fragrances are used by consumer brands to differentiate and innovate. Therefore, fragrances are thought to be a part of science and art [4]. According to consumer research, fragrance is an important factor in deciding a user's choice for any cosmetics. This could be because the ability to sense is intimately linked to the brain's limbic system, which stores feelings and memories. As marketers are well aware, a specific aroma can become strongly associated with product identity and acceptability. [5] The fragrance industry distributes fragrances to consumer goods and fine fragrance producers and retailers, like cosmetics and personal care items. Manufacturers build distinct items that help them stand out and gain a competitive advantage. This can assist increase sales, profits, brand loyalty, and brand recognition.

Cosmetic Fragrances Contents
Pure fragrance compounds or crude extracts acquired through various techniques are typically used as fragrance raw materials. Perfumers combine the raw material to make a fragrance that meets the recognized fragrance requirements, that are typically determined by market research, the sort of cosmetic to which they'll be combined, the impression of cosmetics, the target buyer (age, gender, etc.), individuality, lifestyle, and other factors. Perfumes (also known as fine fragrances) are hydro-ethanolic solutions of essential oils or concentrates that are classified as per the concentration is in the finished products. This classification and perfume contents commonly found in cosmetics are shown in Table 1[6].

Role and Benefits of Fragrance in Cosmetics
Fragrances offer pivotal advantages that are universally recognized, palpable, and appreciated. Cosmetic fragrances are important for providing cosmetics a pleasing perfume because, aside from the design of the packaging, the odour is one of the main things that a buyer notices about the cosmetics. It has been stated that pleasant odours influence product comfortability and effectiveness, as well as play a significant impact in cosmetics' overall evaluation [1,7]. Fragrances are utilized in products to improve the user experience for a variety of reasons. Fragrances can offer a pleasant odour and help cover the smell of some components, as well as make the items more recognized or distinct. According to some studies, fragrances can help to relieve stress and produce well-being by satisfying emotional demands and communicating notions like hygiene, freshness, and tenderness[8].

Fragrances in Cosmetics and Personal Care Products
A large number of personal care products contain fragrance components. The fragrance is found in cosmetics such as shower gels, shampoos, body lotions, and shaving creams to enhance the customer's pleasure as well as their general well-being. For example, fragrant soap can urge consumers to clean their hands more frequently, resulting in improved hygiene. Even unscented items may include fragrances to hide the odour of other components [8]

Fragrances in Cleaning Products
Many household items, including fabric softeners, laundry detergents, room fresheners, carpet fresheners, and dryer sheets include the fragrance [8].

The socio-economic benefits of fragrance [4]
The worldwide fragrance industries contribute enormous economic and social value to suppliers, consumer products makers, retailers, and individuals all over the world.The fragrance business generates value and creates jobs through operations like production, mixing, and research and development (R&D). The fragrance industry's global activities generate €2.5 billion in valuation and provide over 15,000 employments. Employee spending in the fragrance sector generates an additional €0.3 billion in value. Another 11,000 full-time employment are supported by their spending

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in the economy. When totalled, the €2.8 billion in value-added represents 39% of the €7.2 billion in value-added generated by the fragrance sector.

History of Fragrances

The origins of the term "perfume" are "per" which means "through" and "fumum" which means "smoke," implying that the original fragrances were pleasant odours produced by igniting woods, grasses, and other materials. Ancient literature and archaeological digs show that Some of the oldest human societies used perfumes. Ancient humans used spices and herbs including coriander, almond, bergamot, pine resin, and myrtle, along with flowers, in perfumes. The Indus civilization (3500-1300 B.C.) in India had its perfume and perfumery. One of Ittar's first distillations(attar) is described in the Indian Ayurveda scriptures Charaka Samhita and Sushruta Samhita [5,7,9,10]. Perfumery originated in ancient Mesopotamia and Egypt and was developed by the Romans and Persians. Tapputi, a perfume manufacturer stated in a cuneiform tablet from Mesopotamia dating from the 2nd millennium B.C. (2000-1001 B.C.), is thought to be the world's earliest known chemist. According to reports, she distilled calamus, flowers, and oil with other aromatics, then filtered and reused them multiple times [11]. In Pyrgos, Cyprus, archaeologists have unearthed what they believed to be the oldest known scents on the planet, which date back over 4,000 years in 2004 and 2005. At least 60 stills, funnels, perfume bottles, and mixing bowls from an antique perfumery were discovered in a 4,000 m2 (43,000 ft2) "factory." [9].

The Persian scientist Ibn Sina devised the distillation method of obtaining oils from flowers in the early 11th century, which is being used today. He began by experimenting with a rose. Until his innovation, liquid fragrances were made from a powerful mix of oil and crushed plants or petals. Rosewater, on the other hand, was gentler and gained popularity quickly. Later European perfumery and discoveries, especially in chemistry, were greatly influenced by the raw materials and distillation process.The art of fragrance has been practiced in Western Europe since 1221. Fragrances were mostly used by the rich to conceal body odours caused by irregular bathing between the 16th and 17th centuries, and the perfume industry thrived as a result of this patronage [12]. Giovanni Paolo Feminis, an Italian barber, produced Aqua Admirabilis, perfume water that is now known as eau de cologne, in 1693. Aromatic plants were being planted in the 18th century in the grass's region of France, Calabria, and Sicily (in Italy) to supply raw materials to the booming perfume business. The epicentres of European fragrance production and business are still in Italy and France today. The industrial synthesis of aroma molecules like vanillin and coumarin in the late 1800s paved the way for the manufacture of perfumes with fragrances previously inaccessible from natural aromatics [11,13,14].

Market Overview of Fragrance Ingredients

The fragrance industry is an important part of the fragrance value chain, which extends from ingredient suppliers through consumer product manufacturers and retailers. The fragrance industry generates €7.3 billion in sales globally, with downstream Consumer Product Goods (CPG) manufacturers earning €357 billion in manufacturing and retailing revenues (68 % personal care/cosmetics, 9% fine fragrance, and 23 % in-home care/cleaning) [4]. Due to rising consumer expenditure on premium fine fragrances and perfume goods, the Fragrance Ingredients Market surpassed USD 14.5 billion in 2020, with a CAGR of around 7.5 % predicted from 2021 to 2027. The cosmetic industry's rapid growth, combined with the increased inclusion of fragrance compounds in cosmetics and toiletries, as well as fine fragrances and perfumes, is creating advantageous commercial opportunities [15].

Regulatory Aspects for Fragrance Ingredients in Cosmetics
United States of America (USA)

Many of the goods we use daily contain fragrances. Some of these items are regulated by the US Food and Drug Administration (USFDA) as cosmetics. The product is defined as a cosmetic under the Federal Food, Drug, and Cosmetic Act if it is intended to be applied to a person's body to cleanse or beautify it. Perfume, aftershave, cologne, lipstick, conditioner, and skin lotions are examples of cosmetic fragrance products [8,16]. Fragrance products administered to the body for medicinal purposes are classified as drugs by the FDA. For example, if the fragrances

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manufacturer states that it helps "ease muscle aches," "soothe headaches," and "helps people sleep," the fragrance material is regarded as a drug. Under the law, products intended for this type of use are classified as drugs, or as both cosmetics and drugs. Toothpaste with fluoride and makeup with sunscreen are examples of fragrant products that are both cosmetic and drugs. There are plenty of additional products that could incorporate fragrance compounds but they are not used on the body such as dryer sheets, fabric softeners, laundry detergents, room carpets, and fresheners are governed by the Consumer Product Safety Commission (CPSC) [16]. Cosmetic fragrance ingredients should follow the same safety standards as the rest of the cosmetic ingredients. The law does not require approval from the FDA before they can be marketed, but they must be safe for consumers when used as instructed on the label or as people normally do. Cosmetics manufacturers and marketers have a legal obligation to ensure that their products are safe and appropriately labelled. [16]

IFRA and RIFM

The International Fragrance Association (IFRA) has a comprehensive program to evaluate the safety of fragrance compounds. This programme, which has been in effect dating back to 1973, involves a Code of Practice that contains guidelines for good manufacturing practices as well as guidelines for evaluating the safety of fragrance ingredients, including safety criteria that restrict or prohibit the use of specific fragrance materials. The IFRA is in charge of gathering evidence on the safety of certain fragrance compounds and reviewing it to assess their safety within specific use circumstances. The findings of the IFRA safety assessment are documented in the IFRA Code of Practice, which gives essential information to the formulators of fragrances for determining product safety.IFRA publishes its guidance relies on the studies of the Fragrance Materials Research Institute (RIFM), which undertakes scientific studies on fragrance compounds. The RIFM was established in 1966 as a non-profit scientific organization to generate and review fragrance ingredient safety data. The organization collects and analyses scientific data, conducts tests and evaluations, disseminates information, collaborates with government authorities, and promotes consistent fragrance ingredient safety standards [17,18].

In this regard, the RIFM's Expert Panel for Fragrance Safety reviews the actions of the RIFM, as well as the industry and government organization, independently. These three non-profits seek to supply the industry with unbiased information. The RIFM's Expert Panel for Fragrance Safety evaluates the existing scientific research on fragrance constituents, which includes dermatologists, pathologists, toxicologists, and environmental scientists. The International Fragrance Association (IFRA) has developed a Code of Practice based on these findings, which offers scientifically-based guidance for the proper use of fragrance components in products. The Code of Practice, which is implemented by IFRA member companies all over the world, concerns the production and management of all fragrance ingredients for all kinds of uses, especially cosmetics and personal care [19].

However, participation in IFRA is voluntary and the Code of Practice is not legally enforceable, IFRA members must comply with it. According to IFRA statistics, 90 % of the worldwide fragrance business is presently provided by its members. In reality, most cosmetics companies want their perfumes to meet IFRA requirements. New or revised usage restrictions are added to the IFRA Code of Practice regularly [20]. Since January 20th, 2020, a new Amendment was put in force. Within 25 months, all fragrances need to comply with the 49th IFRA Amendment. IFRA Certificates of the 48th Amendment will not be accepted after February 2022[21].

European Union (EU)

Any cosmetic product sold in the European Union must adhere to the EU Cosmetics Regulation 1223/2009, which includes fragrance components [22]. As a result, regardless of IFRA affiliation, every fragrance firm intending to sell its products in the EU has to follow the Cosmetics Regulation. The Cosmetics Regulation is legally binding, unlike the IFRA Code of Practice. In the event of a conflict between the IFRA Code of Practice and the Cosmetics Regulation, the latter must prevail in the EU [20]. Cosmetic products put on the European market must have a product information file (PIF). If a product includes fragrances, the PIF must also include an allergen list as well as an IFRA certificate. An IFRA certificate is a document produced by a fragrance manufacturer certifying that the

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fragrance complies with IFRA criteria and is safe to use. The certification specifies the highest level of fragrance that can be used in specific product categories. The IFRA certificate can be used by the safety assessor to see if the level of fragrance in a particular cosmetic product is within the IFRA standards' limit [23]. IFRA and RIFM are used to identify fragrance ingredients in the United States, Canada, and Europe. This effectively means that the global fragrance industry is self-regulatory [24].

Fragrance Ingredients Safety and Testing
The International Fragrance Association (IFRA) has compiled a list of 3,059 fragrance ingredients. Some of the 3,059 substances have been linked to health issues like allergies, sensitivities, reproductive toxicity, cancer, and according to research.A study in 2016 looked at the effects of fragrances on people's health. According to the research of a random sample of US citizens, 99.1% of people are introduced to fragrant goods at a minimum once every week, either through their use, the use of others, or both. Participants also reported a wide range of health issues related to fragrance exposure, including migraines, asthma, gastrointestinal issues, and cardiovascular issues. The results revealed that a large number of the participants were unaware of the chemicals used in fragrance and if they realized a fragranced product produced pollutant, they would avoid using it [25].

Diethyl phthalate (DEP) is a phthalate that is often used as a fixative and a solvent in fragrances (a chemical that lengthens a fragrance on the body). Neantine, Peilatinol A, and Solvanol are some of its trade names. DEP-containing fragrances have not been determined to be harmful to human health when used in their current form. The Cosmetic Ingredient Review Expert Panel confirmed in 2002 that DEP was shown to be safe for usage in cosmetics.The International Fragrance Association (IFRA) and the Research Institute for Fragrance Materials (RIFM) collaborate to ensure the safety of fragrances in humans. The IFRA standards are used by the fragrance industry to ensure the safe use of fragrance. RIFM has been researching and producing fragrance material safety profiles for more than 50 years[8].Safety testing on fragrances is conducted by RIFM scientists utilizing a four-step method that should be performed before a fragrance can be licensed for use in the products.

Identifying the hazard: Test to see whether the fragrance could trigger an allergic reaction, for example, a rash on the skin.
Dose-response analysis or hazard quantification: Determine how much fragrance presence is likely to have a negative effect.
Exposure assessment: Establish how and how much of each fragrance component will be employed.
Risk characterization: Use consumer products to test the fragrance to assess the level of exposure that is acceptable.

Labelling Requirement of Fragrances in Cosmetic
USA: The label on each container of a cosmetic must declare the name of each ingredient in decreasing order of predominance, apart from that fragrance or flavour certainly stated as fragrance or flavour, according to 21 CFR 701.3(a). [26] If a fragrance component acts as both a flavour and a fragrance, it must be labelled as "flavour and fragrance." A fragrance or flavour's constituent can also be identified by their respective label names.The component or mixture of ingredients that act as a masking agent, i.e., covers the unwanted off-odour of a product without adding a perceptible odour, may be identified by their name(s) or by the term "fragrance." Under 701.3(1)(2)(iii), a masking agent contained in a product at a low concentration, it may be called an incidental ingredient, and hence need not be stated on the label [27]. Under the Fair Packaging and Labelling Act (FPLA), the FDA and the Federal Trade Commission (FTC) oblige the disclosure of ingredients on retail products. This law isn't applicable to compel a firm to divulge "trade secrets." Fragrance and flavour formulae are complicated blends of natural and synthetic chemical compounds, and they're the kinds of cosmetic ingredients that are considered to be "trade secrets." As a result, the FDA allows product makers to group fragrances on the label under the term "fragrance." [28].

EU: The words 'parfum,' 'fragrance,' or 'scent' in the ingredients list, or the name of a specific essential oil, will appear in all cosmetics that contain any fragrance compounds, flavour, or aroma. Fragrance ingredients are

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examined to see if they are likely to induce allergic responses on the skin. There are 26 fragrance components, listed inTable 2 that are more likely to trigger reactions in people. When the concentration of fragrances surpasses 0.001% in leave-on products (e.g., sunscreen) and 0.01 % in rinse-off products (e.g., a soap), they must be listed in the ingredients list alongside the word 'parfum'.This method of labelling cosmetics was launched in 2005 to assist consumers in making educated purchasing decisions, particularly if they have a known sensitivity to a specific scent ingredient. This type of labelling will also assist dermatologists in determining the reasonableness of a patient's reaction. Patients who have been advised to avoid all fragrances must avoid any product containing parfum, perfume, fragrance, aroma, essential oils, or botanical extracts. [29]

Additional fragrance allergen labelling [31]
In 2012, the Scientific Committee on Consumer Safety recommended that, in addition to the current 26, customers be notified of further 61 fragrance allergens on cosmetic product labelling. This new law, if implemented, would have a substantial impact on the way cosmetics are labelled. As a result, the European Commission has proposed three approaches for dealing with this:The present labelling requirement will remain unchanged.Additional fragrance allergens should be labelled per current labelling regulations.Fragrance allergens can be digitally labelled as an alternative to on-pack labelling by using a website, a QR code, or a barcode, or a combination of these.

Fragrance Allergies and Sensitivities
Allergens are chemicals that cause the immune system to react. Irritation, redness, and even hives or swelling are common symptoms of skin allergens. Even while unfavourable skin reactions to cosmetics and scents are defined as sensitivity rather than allergy, the difference is negligible to the sufferer. Fragrance/cosmetic allergen labelling may vary widely because individual reactions might vary greatly and the reaction itself does not approach the severity of food allergens [32]. Even if some components in cosmetics, food, or other items are safe for most people, a small number of people may be allergic or sensitive to them. Even though some chemicals do have the possibility to trigger adverse reactions, they can be used in small amounts in the products. A safety evaluation can be undertaken using a method called Quantitative Risk Assessment (QRA) to evaluate permissible usage limits of fragrance components in a range of consumer products[33]. Primary contact allergy prevention from consumer items is based on exposure management. Secondary prevention is ensured through product labels [34]. The FDA in the United States, for example, notes allergic reactions to scents, but instead of enforcing label declarations of individual components, they advise consumers with fragrance sensitivity to seek fragrance-free products.

Transparency of the Fragrance Ingredients
All of us do have the right to be informed and comprehend what's contained in our products. The Personal Care Products Council (PCPC) and its member have a strong record of supporting consumer safety and transparency. In the present advanced world with a consistent inundation of data accessible, individuals have grown accustomed to getting easy accessibility to goods details. Because of this, IFRA fostered the IFRA Transparency List, a web-based database of all fragrance components utilized in products around the world. The website records the components used in manufacturing the products. Several PCPC member firms have adopted many voluntary initiatives to improve transparency and disclose details of fragrance to customers and other parties involved and incorporated the "Voluntary Contact Allergen Disclosure Guideline" to the PCPC Consumer Commitment Code. Consumers can utilize the contact information on product labels to contact the company if they have queries about specific items or ingredients [35,36].

CONCLUSION

Many products have the word "fragrance" on the label, but only some specify the elements which constitute a "fragrance." Consumers are unable to obtain a complete list of components in their products due to this lack of openness. Whereas the majority of fragrance ingredients aren't labelled, we know that few have been known to cause reproductive and developmental damage, cancer, sensitivities, and allergies. There is a requirement for more grounded guidelines, further exploration, and more prominent straightforwardness. The IFRA Code of Practice and

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country-specific rules on the use of fragrance compounds may not every time align, and lawfully obligatory cosmetics rules are frequently further limited than the IFRA Code of Practice. Adopting the IFRA standards, even though designed to assure consumer safety, it's doesn't completely ensure a fragrance's regulatory requirements in the nation, as fragrance companies hoping to market their goods also need to comply fully with the country's specific cosmetics laws.

Declaration Conflict of Interests
The authors declare that there is no conflict of interest for the publication of this manuscript.

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Table 1: Perfume contents commonly present in cosmetics.

S. No	Cosmetic product	Approximate content (%)
1.	Baby cologne	1–2
2.	Cologne	2–3
3.	Eau de cologne	3–4
4.	Eau fraiche	4–5
5.	Eau de toilette	5–15
6.	Eau de parfum	15–20
7.	Parfum	20–40
8.	Other products	
	• Skincare products	0.01–0.5
	• Haircare products	0.01–1
	• Bath preparations	0.1–3

Table 2: List of ingredients that may trigger reactions in humans [30]

S. No.	INCI Name	Origin	Sources
1.	Alpha-Isomethyl ionone	Synthetic	N/A
2.	Amyl cinnamal	Synthetic	N/A
3.	Amylcinnamyl alcohol	Synthetic	N/A
4.	Anise alcohol	Natural or synthetic	Honey, essential oils of Anise, Tomatoes, Tahiti Vanilla
5.	Benzyl alcohol	Natural or synthetic	Peru Balsam, Tolu Balsam, Essential oils of Jasmin, Apricot, Almond, Apple.
6.	Benzyl benzoate	Natural or synthetic	Peru Balsam, Ylang-Ylang, Tolu Balsam, Essential oils of Jasmin.
7.	Benzyl cinnamate	Natural or synthetic	Tolu Balsam, Copahu, Peru Balsam
8.	Benzyl salicylate	Natural or synthetic	Propolis
9.	Butylphenyl methylpropional	Synthetic	N/A
10.	Cinnamal	Natural or synthetic	Essential oils of Nutmeg, Patchouli, Cinnamon,

			hyacinth
11.	Cinnamyl alcohol	Natural or synthetic	Hyacinth
12.	Citral	Natural or synthetic	Essential oils of Orange peel, Lemon, Eucalyptus
13.	Citronellol	Natural or synthetic	Essential oils of Ceylon, Lemongrass.
14.	Coumarin	Natural or synthetic	Angelique, Berce, Flouves, Sweet clover, Woodruff,
15.	Eugenol	Natural or synthetic	Essential oils of Ceylon, cinnamon Bay (Myrcia acris), Cistus Avens, Laurel, Clove, Allspice.
16.	Farnesol	Natural or synthetic	Essential oils of Tolu Balsam, Lime tree, Neroli, Ylang-ylang, Rose
17.	Geraniol	Natural or synthetic	Orange, Rose oil, Thyme, Verbena, Myrtle, Melissa, Palma Rosa, Apple, Neroli, Apricot, Nutmeg,
18.	Hexyl cinnamal	Synthetic	N/A
19.	Hydroxycitronnellal	Synthetic	N/A
20.	Hydroxyisohexyl 3 cyclohexene carboxaldehyde	Synthetic	N/A
21.	Isoeugenol	Natural or synthetic	Essential Oils of Ylang-ylang, Ceylon, Citronella,
22.	Limonene	Natural or synthetic	Essential oils of Verbena, Dill, Common, Orange, juniper, Lemon, Niaouli, Neroli.
23.	Linalool	Natural or synthetic	Essential oils of Pine, Peppermint, Marjoram, Laurel, Sour orange, Thyme Lemon, Lavender orange.
24.	Methyl 2-octynoate	Synthetic	N/A
25.	Evernia prunastri	Natural	Oak moss extract
26.	Evernia furfuracea	Natural	Tree moss extract

Figure 1: Fragrance ingredients market size [15]

REVIEW ARTICLE

A Mini Review on Possible Diagnosis Errors in COVID 19

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ABSTRACT

Coronavirus created a worldwide havoc through its outbreak in Wuhan, China in 2019 Dec and it is continuously spreading all over the world. This pandemic disease has named as novel coronavirus disease 2019 (COVID-19) which causes severe acute respiratory syndrome coronavirus 2 (SARS-CoV-2) in their hosts. The current gold standard method for the diagnosis of SARS-CoV-2 infection is RT-PCR of patients respiratory specimens, however certain other diagnostic techniques such as serological techniques are implemented too. Even though there are various means of diagnosing the virus, at the same time the techniques also may face some vulnerabilities which may yield errorsdue to many preanalytical issues such as inadequate procedures for collection, identification problems, handling, collection of inappropriate or inadequate material, transport and storage of the swabs, presence of interfering substances, manual errors, as well as specific aspects such as sample contamination etc. Errors in such fields may contribute to jeopardize the diagnostic accuracy.

Keywords: COVID-19, Corona virus, Clinical errors, Diagnosis, Serological techniques,; Nucleic Acid test, Reverse transcription polymerase chain reaction (RT-PCR).

INTRODUCTION

Coronavirus belongs to a family of viruses that cause illnesses through a pneumonic condition in an individual. The symptoms range from mild to severe. The three most common illnesses by these viruses recorded were: SARS, MERS and COVID19. SARS in full form severe acute respiratory syndrome is a respiratory illness discovered in China in 2002.However there haven't been any new cases reported under this virus since 2002. MERS (Middle East respiratory syndrome) is a severe respiratory illness first reported in Saudi Arabia in 2012 and then spreaded to 27 countries. And recently, the COVID-19 (Coronavirus disease 2019), erupted in 2019 in Wuhan, China creating a pandemic situation throughout the world. The communicability of this virus is so high, so to avoid this spreading, the whole globe was put into lock down and precautionary measures and diagnostic measures were placed. The communicability of the virus possesses a grave threat as the rate of infection supersedes the diagnostic measures in a vast magnitude. In such scenario, errors in clinical diagnostic are an important step which should be taken care of.

Types of Anticipated Diagnostic Errors
COVID-19 represents the respiratory illness which requires confirmatory tests, but the required tests may not be available or produce a false-negative result leading to an error termed as "Classic.". There is a wide local and regional variation in the type of tests, availability of tests, as well as accessibility of information regarding test performance characteristics or diagnostic yield [1]. Test results that yields false negatives or tests that are not performed due to non-availability of resources can lead to delay in diagnosis of the disease and the further spreading of the pandemic. Tests are even more important when patients present unusual / non-respiratory symptoms. Both predominantly olfactory [2] and gastrointestinal manifestations [3]have now been described along with multisystem inflammatory syndromes which are different from the respiratory symptoms as discussed earlier. The failure to recognize a typical presentations either because of testing problems or knowledge gaps, could lead to an error termed as "Anomalous" which overlook the underlying COVID-19 diagnosis. Mislabeling patients who do not have COVID-19 as having the disease is another error emerging in the pandemic particularly in those exhibiting respiratory symptoms. This error is common when there is absence of testing in a saturated health system with limited capacity to test or treat. "Anchor," is the term used to define the type of error which can introduce various risks such as missing other respiratory infections (E.g. pneumonia) and non-respiratory conditions. In another scenario, where the patients with known COVID-19, a second underlying condition may be missed, such an error is termed "Secondary." The collateral effects of the COVID-19 pandemic are also emerging, for example, a patient with symptoms of the acute conditions may be un willing to visita cute care for evaluation because of infection risk, and such error is termed as "Acute Collateral". The "Chronic collateral" is another type of error caused due to delayed diagnosis of ambulatory conditions, cancelled appointments or elective procedures. Furthermore there can be strain error where there is a possibility of missing or being delayed in diagnosing a different condition due to hospitals being overwhelmed .This may potentially limits the time and attention that the clinicians spend on non-COVID-19 patients. Physicians, including surgeons, pediatricians, and radiologists, have been "redeployed" into acute care medical specialties. Cognitive errors increase when clinicians in new roles face unfamiliar situations and disease manifestations. Though the clinicians may be highly experienced previously, they may have insufficient skills and experience in their new roles which not makes them feel comfortable in asking for guidance (4). Lastly, clinicians are increasingly using intermediary mechanisms, such as PPE and telemedicine technologies, which reduce room entries and also reduces the ability of well-trained clinicians to the effective histories, performing physical exams, and and monitoring symptoms. In fact, infection prevention and isolation has been shown to put patients at risk of preventable adverse events in hospitalized patients [5]. Which leads to a type of error termed as "Unintended".

Diagnostic approaches towards COVID patients:
The symptoms of coronavirus arises when people has travelled to the areas of the virus outbreak or have been in contact with anyone traveling from those areas. The symptoms in general are: fever, cough, shortness of breath, sore

throat and headache. However the COVID19 specific symptoms are also fever, cough and shortness of breath. The symptoms of COVID-19 patients are quite milder than SARS and MERS. Various diagnostic approaches are put forth to test COVID-19.The basic approaches which are in usages are discussed below along with their possible flaws.

Serological Diagnosis
(a)Antibody test are similar to a blood test, which involves the use of a few blood samples to determine whether the human body has antibodies for coronavirus. Antibodies are used produced by the immune system to identify and neutralise foreign objects or pathogens. Antibody test is a low cost diagnostic test when compared to the other tests and gives result in 20-30 minutes.

(b)Antigen detection test Interpretation of serology tests may also depends on antigen specificity, which can be proteins, polysaccharides or lipids. Various viral antigens such as spike protein, nucleocapsid and receptor binding domain are being used to detect antibodies for SARS-CoV-2. Based on this principle Antigen-based tests are used to diagnose COVID-19. The sensitivity of these tests might be expected to vary from 34% to 80% [6]. Hence testing for antibody can be misleading and hence have higher chances of diagnostic errors as the tests work depends on several factors such as the time from onset of illness, the viral load in the specimen, the quality of the specimen collected, processing of the sample and the precise formulation of the reagents in the test kits.

(c) Host antibody detection test Another rapid diagnostic test for COVID-19 is the test that detects the presence of antibodies in the blood samples that are believed to have been infected with COVID-19[7-11].Antibodies are produced over days to weeks after infection with the virus. But a point to be noted is that the strength of antibody response varies from individual to individual pertaining to several of their factors such as age, nutritional status, severity of disease, and certain medications or any viral load that is supressing the immune system[11-13].These factors can be responsible in altering the viability of the results. Even in some cases where people were confirmed with the positive COVID-19 disease (molecular test), weak or negative antibody responses have been reported[11,12,14].Studies suggest that the majority of patients develop antibody response only in the second week after onset of symptoms[7,11,12,15-19]. These facts may lead to a conclusion that a diagnosis of COVID-19 infection based on antibody response will often only be possible in the recovery phase, which will be too late for the clinical interruption of disease and transmission have already pass to another individual. Another issue faced by antibody detection tests is targeting COVID-19 may also cross-react with other pathogens, including other human coronaviruses[12,20,21] and give false-positive results.

NAT for COVID-19 NAT stands for Nucleic Acid Test. It requires a nasal and throat swab. The cost of NAT is lower compared to that of RT-PCR Test and takes about an hour. The test is simplistic and requirement of highly experienced staffs can be listed out from the requirement. Moreover multiple tests upto 4 can be done in a same allotted time, so coverage for more individual is an advantage seen in this technique.

RT-PCR Tests For COVID-19 Among the two tests mentioned earlier RT-PCR is the most precise technique and gives us the accurate result as the test is conducted against the virus directly. The flaws is that the entire process is lengthy as from collecting the sample to delivery of report takes a day along with its high cost. Another condition put forwarded by this test is that it requires highly trained personals to conduct this test. These conditions prevailing in the background gives us the outlook of time efficiency which we are unable in competing against fast spreading virus.

Lateral flow assay Another diagnostic approach has been put forth by the Canadian firm called Sona Nanotech have been attempting to push a completely different kind of rapid screening test for Covid-19 to market. It's a quick-response lateral flow assay, and the firm states its test to produce results in five to 15 minutes, cost less and the ability to administer the test by untrained individuals. Lateral flow assays have a broad spectrum of applications and can test a variety of samples starting from urine samples to blood, saliva, sweat, serum, and many other bodily fluids. All these tests are designed to identify the presence of a specific biological marker. It isn't a PCR test which

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requires a swab, a wait and specialist analysis to yield a result. Many antibody tests rely on lateral flow, but it isn't one of those either as the test will directly detect the Covid-19 virus.This technology should make it suitable for in-home testing and monitoring, to help identify if patients need treatment in a clinical facility. It is also believed that this technique will be able to verify if people are ready for release from quarantine and to screen individuals prior to entering closed public venues.

CONCLUSION

The COVID-19 emergency which has now become pandemic, is remarkably harnessing the usage of laboratory resources for diagnosing SARS-CoV-2 infections through various diagnostic tools and technique. Unfortunately, it cannot be excluded that, the quality of tests for detecting the virus and in offering accurate results could be jeopardized by a number of pre analytical and analytical factors. The factors responsible for an error is not limited to the coronavirus but infact can run through any other disease as well; however the situation is worsened up due to the current status of the world being in a pandemic situation. Apart from the technical error, there is higher probability of lessening the clinical error and the aforesaid could be in a better shape by finding our more reliable source of diagnosis. The more accurate diagnostic tool and technique need to be set forth along with an individual responsibility in margining the human errors.

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Fig.1. User-Friendly Nomenclature of Diagnostic Errors Anticipated in the COVID-19 Pandemic	
Nomenclature	**Description**
Classic	Missed or delayed COVID-19 diagnosis in a patient who presents with respiratory symptoms
Anomalous	Missed or delayed COVID-19 diagnosis in a patient who presents with non-respiratory symptoms
Anchor	Missed or delayed non–COVID-19 diagnosis because it was assumed to be COVID-19
Secondary	Missed or delayed non–COVID-19 or secondary diagnosis in a patient being treated with known COVID-19 disease
Acute Collateral	Delayed diagnosis of acute non–COVID-19 diagnosis because patients are not coming in for evaluation because of the infection risk
Chronic collateral	Error which may be caused due to delayed diagnosis of ambulatory conditions due to cancelled appointments or elective procedures.
Strain	Possibility of missing or delaying in diagnosing a different condition due to hospitals being overwhelmed
Unintended	Any missed or delayed diagnosis because of less direct interactions, including rapid increase of telemedicine and PPE

RESEARCH ARTICLE

High Carveol Yielding Essential Oil Sourced Out From Wild *Cymbopogon giganteus* (Hochst.) Chiov.

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ABSTRACT

Wild genotype of *Cymbopogon giganteus* (Hochst.) Chiov. was collected from Jnana Bharathi campus, Bangalore. Plant was identified via; DNA barcoding studies and the sequence was deposited in NCBI GenBank under the accession number OK094426. Plant was subjected to hydro distillation for the isolation of essential oil using Clevenger apparatus. Quantitative analysis of the essential oil was performed using GC-MS. 63 compounds were identified in the essential oil representing 100% of the total oil. The major constituents in the oil are Carveol (28.07%), Iso-carveol (22.28%), cis-carveol (5.79%), p-menthane (18.15%), cis-p-mentha-2,8-dien-1-ol (5.93%) and trans-p-mentha-2,8-dienol (12.22%). Exploration of wild plants would lead to significant economically valued pharmaceutical compounds producing species.

Keywords: Wild *Cymbopogon giganteus*, Essential oil, DNA Barcoding, Monoterpenoids, Sesquiterpenoids, GC-MS

INTRODUCTION

Aromatic plants are mainly characterized by the presence of aromatic compounds[1]and possess odorous volatile substances which exist as essential oil, gum exudate, balsam and oleoresin in one or more parts, namely, root, wood, bark, stem, foliage, flower and fruit[2].Plants are serving as source of folk medicine since time immemorial and they are used as modern medicines, nutraceuticals, food supplements, pharmaceutical intermediates and chemical entities for synthetic drugs due to presence of various types of bioactive phytochemicals, essential mineral elements and other pharmacological properties. Bioactive compounds like alkaloids, flavonoids, terpenoids, tannins and phenolic compounds are the basis for the medicinal value of plants that produce a definite physiological action on the body [3]. The genus Cymbopogon is of significant interest due to its commercially valuable essential oils and is widely used in functional food as well as in traditional medicine [4]. The genus, comprises of about 180 species, subspecies, varieties and subvarieties[5] and belongs to the Family Poaceae, which is listed to have about 660 genera and 9000 species[6]. The genus is native to warm temperate and tropical regions of the Old World and Oceania[5]. Essential oils of plants belonging to the Cymbopogon genus can be sought for various properties such as their insecticidal, analgesic, anti-inflammatory, antioxidant, hypotensive, anti- leishmaniasis, anti-microbial, antiplasmodial, anti- acetylcholinesterase, anti- trypanosomal, anti-nociceptive and anticonvulsant potentials[7].

C. giganteus is applied in folk medicine against various diseases such as skin disorders (decoctions of leaves and flowers from scorpion bites), mental illness, broncho-pulmonary affections, bilharzia, headaches, jaundice, cold, conjunctivitis, migraine, dermatoses, rheumatic pains, childhood coughs and hepatitis. Essential oil of *C. giganteus* obtained by hydrodistillation of its leaves shows good antibacterial and anti-inflammatory properties and is sold for these activities [8]. They are also used for febrifuge, pulmonary, antiiceteric disinfectants and antimalarial properties [9]. A mixture of boiled water extracts of *C. giganteus* has strong effect against chloroquine resistant Plasmodium [10]. Aqueous decoction of stems with leaves of *C. giganteus* and *Ocimum basilicum* is used to treat sickle cell disease and decoction of only their leaves, in the treatment of drepanocytosis. The decoction of their leafy stems is used for calming epileptic seizures also. Essential oil is used against boils, stomach pain and toothache [11]. *C. citratus* and *C. giganteus* are traditionally used in Congo for their anti-ulcerative potential. In Burkina-Faso, fresh roots decoctions are used against toothache, gingivitis and sores in mouth, tongue and on the lips. In Benin, the whole plant is burnt for its repellence property against mosquitoes [7].

C. giganteus is a widely studied *Cymbopogon* species which has a great reputation in the W. African pharmacopoeia as a panacea [12]. *C. giganteus* is an aromatic perennial grass, possessing a rhizome-bearing stem[10]with slightly aromatic glaucous leaves [7]and can grow up to 2-3 metres [10]. It is widely distributed in the Asian and African tropical Savannahs [13]. The plant is a loosely tufted herb with robust culms and leaf blades are linear to narrowly lanceolate [14] and the lower most inter node and pedicel are connate [13]. The plant possess dense panicles and flower appears throughout the year [7]. Essential oil of *C.giganteus* contains diverse bioactive compounds and also possess phytochemical and antimicrobial activities thereby finding many applications in folk medicine. Worldwide demands of essential oils of *C. giganteus* has escalated during the past few years [15]. Gas chromatography Mass Spectrometry (GC-MS) applied for the analysis of aromatic and medicinal plants has proved to be a valuable method to analyse the essential oil, alcohols, acids, esters, alkaloids, steroids, amino and nitro compounds [16]. In the present work, the wild *C. giganteus* was explored to estimate the quality and quantity of its essential oil. As a result, a thorough essential oil characterization was carried out.

MATERIALS AND METHODS

Plant Collection
Fresh herbage including rhizome, culms, leaves and inflorescence from wild *Cymbopogon sp.* growing in Bangalore University, Jnana Bharathi campus were collected for the study. Bangalore University is located on the western side of Bengaluru city, sprawling 1100 acres of land with an altitude of 900m, 12º57ˈ02ˈ N latitude, 77°30ˈ13ˈ E longitude

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and time zone – IST (UTC/GMT+5:30 hrs. Temperature of 27° C, precipitation of approx. 3%, wind flow of 8.1 miles/hr, rainfall levels of approx. 0.2 and humidity of approx. 6% was recorded on 5th January, 2021 (the day of collection).

Plant Identification
Isolation Of Total Cellular DNA And Primer Designing for Barcode Loci Amplification
Fresh and young leaves of the wild plant were taken and subjected to total extraction of cellular DNA using CTAB method[17]. The corresponding gene sequences of the genus Cymbopogon were retrieved from NCBI Gen-Bank data domain for precisely designing the specific primers for the amplification of three barcoding loci, i.e., *rbcL, matK* and ITS1 and 2 spacers. PCR primer pairs were mapped out from the conserved regions using software primer 3.0 (version 0.4.0) [18].

Barcode Amplification, Sequencing, Validation and Data Analysis
Two chloroplast loci (rbcL, matK) and one nuclear DNA locus (ITS region) of the isolated DNA from the fresh young leaves were amplified using the primers that were designed. PCR reaction mixture contained the template DNA, buffer, MgCl$_2$, dNTPS, designed primer and DNA polymerase. The PCR program that was set involved 35 cycles, each cycle starting from an initial stage of denaturation at 94°C for 5 mins, followed by an annealing stage at 60°C for 1 min, extension stage at 72°C for 2 mins and final extension at 72°C for 10mins. The PCR products were purified and sequenced [18]. Sanger sequencing of amplicons was done using BDT v3.1 Cycle sequencing kit on Abi 3730xl Genetic Analyzer. Annotation softwares were used to annotate the sequenced data. Validation of the designed primers and sequenced data was done by repeating the experiment twice from the starting DNA isolation step to the sequencing step. The PCR products were also subjected to 1.6% agarose gel for the visualization of the amplified products. The gel was pictured with a Gel Doc XR+ (Biorad). Annotated contig barcode sequences were subjected to BLASTn (NCBI domain) for verification and were finally submitted to GenBank of NCBI. The DNA sequences were aligned automatically using the program CLUSTALW in MEGA 6.0 and constructed NJ derived phylogenetic tree.

Essential Oil Extraction
Fresh plant was collected from the experimental site. Leaves, inflorescence, stems and roots were separated and washed under tap water to remove dust particles and were then dried at ambient temperature for two days in the laboratory. Dried parts were cut into small pieces which were then used for the extraction of essential oils. The plant materials were subjected to hydro distillation using a Clevenger apparatus for 3 hrs. The oil was dried over anhydrous sodium sulphate and was then stored in sealed vials in a refrigerator at 4ºC until analysis.

GC-MS Analysis Of Essential Oil
Essential oil analysis of wild *C. giganteus* was done using Gas chromatography mass spectrometry. Investigation of essential oil chemical compounds was carried out using Shimadzu GCMS QP- 2010 Plus equipment with the condition of tool specifications as follows: Rtx- 5ms capillary column type of column length 30 metres, column diameter of 0.25mm, column thickness of 0.25 μm; injector temperature of 250ºC, carrier gas Helium with flow rate of 0.7ml/min. an injection volume of 0.1μl sample was employed in a split mode at a split ratio of 1:100. The GC condition had an oven temperature programmed at 40ºC maintained for 2 mins. The temperature was then gradually increased with rates of 5ºC/min up to 280ºC for 2 mins and 20ºC/min up to 300ºC with a hold time of 2 mins. 280ºC for interface temperature was maintained. The phytocompounds were identified by comparison of mass spectra with the National libraries (NIST-5).

RESULTS AND DISCUSSION

Plant Identification
DNA barcoding studies were carried out to identify the species of wild Cymbopogon collected from Jnana Bharathi campus. Out of three loci (rbcL, matK and ITS spacers 1and 2), only rbcL loci was amplified successfully and

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evolutionary analysis was conducted in Clustal Omega using Neighbour-Joining method. The percentage of replicate trees in which the associated taxa clustered together in the bootstrap test (1000 replicates) are shown next to the branches. The tree is drawn to scale, with branch lengths in the same units as those of the evolutionary distances used to infer the Phylogenetic tree **(Fig 1)**. Evolutionary distances were computed using the Maximum Composite Likelihood method and are in the units of the number of base substitutions per site. Phylogeny indicates that the studied plant sample is very closely grouped under clad of *Cymbopogon sp.* This result supports study of NCBI BLAST leading to confirmation of the species as *Cymbopogon giganteus* and was submitted the same in NCBI GenBank under the accession number of OK094426.

Essential Oil Characteristics
In present study, 100g of the dried herbage sample yielded approximately 0.05ml of essential oil. The essential oil was found to be pale yellow coloured viscous fluid and was characterized by a distinct, powerful, fresh, fruity, floral-herbal aroma.

Essential Oil Analysis
GC-MS analysis identified various compounds such as hydrocarbons, esters, steroids, terpenoids, fatty acids and many other functional compounds. The study on essential oil categorized 63 out of 63 compounds into different chemical classes based upon their structural details **(Fig 2).** The composition of the oil was largely dominated by monoterpenoids (90.5%) followed by oxygenated monoterpenes (4.04%), sesquiterpenoids (4.03%), hydrocarbons (13.37%) including aliphatic hydrocarbons (0.75%), cyclobutanes (11.61%), cyclohexanes (0.20%) and cyclic hydrocarbons (0.81%), azareenes (7.61%), steroids (2.38%), benzofuran (2.23%), fatty acid esters (2.02%), cycloketones (1.82%), saturated fatty acids (0.90), ketals (0.42), epoxides (0.41), alkyl phenyl ketones (0.34), fatty alcohol (0.16%) and diterpenoids (0.14%). The major monoterpenoid proportion in the essential oil mostly contained Carveol compounds (28.07%) and oxygenated p-menthane derivatives such as cis-p-mentha-2,8-dien-1-ol and trans-p-mentha-2,8-dienol (18.15%). 17 out of 63 compounds were identified as higher proportion compounds **(Table 1)** while the remaining 46 compounds were categorized under trace compounds. The compounds showing maximum area percentage were Carveol compounds (Isocarveol and cis-Carveol)-28.07%, p-menthane compounds (cis-p-Mentha-2,8-dien-1-ol and trans-p-Mentha-2,8-dienol)-18.15%, 1,2-Diisopropenylcyclobutane-11.61%, 1,4-Methanopthalazine, 1, 4, 4a, 5, 6, 7, 8, 8a- octahydro-9, 9- dimethyl-, (1alpha, 4alpha, 4aalpha, 8aalpha)- 7.61% and D-Carvone-4.66%. The compounds found in trace amounts included Limonene diepoxide, Limonene 1,2-epoxide, L-Perillaldehyde, Geranyl acetate, Camphene, etc. There were also some characteristic compounds reported in the oil, such as p-cymenene, 1,4-Methanophthalazine,1,4,4a,5,6,7,8,8a-octahydro-9,9-dimethyl-, (1alpha,4alpha,4aalpha, 8aalpha)-, caryophyllene oxide, humulane-1,6-dien-3-ol, sativene and cyclosativene etc. The biological activity of some of the terpenoid compounds found in wild *C. giganteus* essential oil as reported by numerous researchers in the earlier studies has been listed **(Table 2).** It was found that essential oil from wild *C. giganteus* contained greater proportions of carveol compounds with reference to other research studies.[10],[13],[19],[20]

Biosynthesis of Terpenoid Compounds
In present work, biosynthesis of terpenoid compounds via the essential oil pathway was determined based on structural details of the compounds. Monoterpenoids were positioned in MEP pathway (Methylerythritol 4-phosphate) **(Fig 3)** and sesquiterpenoids in MVA (Mevalonate) pathway (Fig4). Biosynthesis of monoterpenoids takes place from Geranyl diphosphate (GPP) as the precursor in chloroplast of the plant cells by the MEP pathway. In fig 3, GPP leads to the formation of 5 more precursors required to form other compounds. Geraniol is one of those precursors formed by the action of the enzyme Geranyl- diphosphate diphosphatase (GDD) of class Hydrolases, reaction aided by a water molecule and release of diphosphate group. Geraniol then forms Geranyl acetate using Alcohol O- acyltransferase (AT) as the enzyme, belonging to the class Transferases. The second compound formed from GPP is (+)- Camphene by the action of (+)- Camphene synthase of class Lyases. The reaction occurs with the release of diphosphate. (+)- Bornyl diphoshphate is the third precursor formed by (+)- Bornyl diphosphate synthase of class Lyases. It then further leads to the formation of (+)-Borneol, reaction aided by the addition of a water

molecule. The enzyme for this reaction is not yet identified but it is of class Phosphoric monoester hydrolases. (+)-Borneol then uses acetyl CoA as the cofactor leading to the formation of Bornyl acetate and CoA. The reaction is catalysed by Monoterpenol O- acetyl transferase (MOA). The fourth precursor formed from GPP is γ- terpinene with the release of diphosphate. γ-terpinene synthase, of the class Lyases catalyses the reaction. γ-terpinene, ultimately forms thymol, carvacrol and p-cymene (formed as the side product). The enzyme demonstrated for this reaction is Cytochrome P_{450} monooxygenases [21]. p-cymene may then give rise to dehydro-p-cymene, with the removal of a hydrogen molecule. The fifth precursor is 4S-Limonene, formed from GPP with the release of diphosphate molecule and reaction is catalysed by (4S)- Limonene synthase of class Lyases. 4S- Limonene may undergo epoxidation and form Limonene diepoxide. 4S- Limonene, with the addition of a water and an acceptor molecule can also lead to the formation of cis-p-Mentha-2,8-dien-1-ol and trans-p-Mentha-2,8-dienol with the release of a reduced acceptor (reaction may be catalysed by the enzyme Limonene dehydrogenase, belonging to the class of Oxidoreductases). 1,3,8-p-Menthatriene can also be formed from 4S-Limonene. It also leads to the formation of Limonene 1,2-epoxide, reaction catalysed by Limonene 1,2-monooxygenase (class oxidoreductases). The reaction is carried out in the presence of oxygen and NADPH+H$^+$ as cofactor, thereby releasing NADP$^+$ and H_2O. Limonene 1,2- epoxide may form Isocarveol. 4S- Limonene also results in the formation of (-)-cis-Carveol (enzyme not yet identified but belongs to the class of oxidoreductases), (+)-trans-Carveol (enzyme catalysing the reaction is Limonene 6-monooxygenase of class oxidoreductases) and Perillyl alcohol (enzyme is Limonene 7-monooxygenase of class oxidoreductase). In case of both (+)-trans-Carveol and Perillyl alcohol, reaction takes place in the presence of a reduced [NADPH-hemoprotein reductase] and dioxygen molecule, thereby releasing an oxidised [NADPH-hemoprotein reductase] and water molecule. Finally, (+)-trans-Carveol results in (+)-Carvone formation using NAD$^+$ as cofactor, thereby releasing NADH+H$^+$. Enzyme involved is (+)-trans-Carveol dehydrogenase of class Oxidoreductases and Perillyl alcohol, in the presence of NAD$^+$, produces Perillaldehyde and NADH+H$^+$. Enzyme involved is Perillyl-alcohol dehydrogenase, belonging to the class Oxidoreductases.

Biosynthesis of sesquiterpenoids via MVA pathway takes place in cytosol of the plant cells. It begins with (2*E*,6*E*) Farnesyl pyrophosphate (FPP) as the precursor molecule. FPP gives rise to (+)-Sativene, Cyclosativene and Beta-Caryophyllene. (+)-Sativene synthase of class Lyases carries out the formation of (+)-Sativene whereas the enzyme for the synthesis of Cyclosativene from FPP is not yet identified but it belongs to the class Lyases. Beta-Caryophyllene is produced by the action of (-)-Beta-Caryophyllene synthase (belongs to the class Lyases). Diphosphate is released in this reaction. Then Beta-Caryophyllene may undergo oxidation to produce Beta-Caryophyllene oxide.

CONCLUSION

Cymbopogon giganteus has achieved a significant position in health management system of most of the developing countries due to its wide applications in several diseases [8]. The plant is immensely used as perfumery and flavouring agent. In the present study, *C. giganteus* was explored from wild field by DNA barcoding. The essential oil obtained from wild *C. giganteus* showed variations in the composition compared to the counter cultivar varieties. Changes in the chemical profile could be associated with abiotic and biotic factors [22]. The terpenoid compounds of the essential oil were traced in MEP and MVA pathways via KEGG and METACYC and further the biological activities of the compounds were recorded which showed the wide applicability of the wild *C.giganteus* in various perfumery and pharmaceutical industries[23].

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Table 1 – Compounds found in high proportion in wild *C. giganteus* essential oil

Sl no.	Chemical class	Area %	Retention time	Molecular formula	Molecular weight
1.	Isocarveol	22.28	17.602	$C_{10}H_{16}O$	152.23
2.	trans-p-Mentha-2,8-dienol	12.22	15.559	$C_{10}H_{16}O$	152.23
3.	1,2-Diisopropenylcyclobutane	11.61	12.630	$C_{10}H_{16}$	136.23
4.	**1,4-Methanophthalazine, 1,4,4a,5,6,7,8,8a-octahydro-9,9-dimethyl-, (1alpha,4alpha,4aalpha,8aalpha)-**	7.61	20.891	$C_{11}H_{18}N_2$	178.27
5.	cis-p-Mentha-2,8-dien-1-ol	5.93	15.921	$C_{10}H_{16}O$	152.23
6.	cis-Carveol	5.79	17.829	$C_{10}H_{16}O$	152.24
7.	D-(+)-Carvone	4.66	19.111	$C_{10}H_{14}O$	150.22
8.	1,3-Bis-(2-cyclopropyl,2-methylcyclopropyl)-but-2-en-1-one	3.33	21.556	$C_{18}H_{26}O$	258.4
9.	Bicyclo[2.2.1]heptan-3-one, 6,6-dimethyl-2-methylene-	2.35	18.532	$C_{10}H_{16}O$	152.23
10.	3,9-Epoxy-p-mentha-1,8(10) diene	2.23	17.417	$C_{10}H_{14}O$	150.21
11.	Androst-1-en-3-one, 4,4-dimethyl-, (5.alpha.)-	1.54	39.632	$C_{21}H_{32}O$	300.5
12.	(2S,4R)-p-Mentha-[1(7),8]-diene 2-hydroperoxide	1.31	22.602	$C_{10}H_{16}O_2$	168.23
13.	Bicyclo[3.3.0]oct-2-en-7-one, 6-methyl-	1.28	16.776	$C_9H_{12}O$	136.19
14.	(-)-Caryophyllene-(II)	1.04	40.413	$C_{15}H_{24}$	204.31
15.	Caryophyllene oxide	1.03	27.801	$C_{15}H_{24}O$	220.35

Table 2: Biological Activity of Some Terpenoid Compounds from wild *C. giganteus* essential oil

Sl. No.	Compounds	Bioactivity	References
1.	Camphene	Antibacterial, antifungal, anticancer, antioxidant, antiparasitic, antidiabetic, anti-inflammatory and hypolipidemic activities, anti-leishmanial, hepatoprotective, antiviral, and anti-acetylcholinesterase inhibitory activities.	24
3.	trans-p-mentha-2,8-dienol	Antimicrobial activities	6
4.	Limonene 1,2-	Potent antinociceptive and anti-tumoral activities, antiparasitic,	25

	epoxide	antimalarial, anti-cancer,anti-microbial.	
5.	cis-Carveol	Strong cytotoxic activities, anti-tumoral activities, chemo preventive against mammary cancer, neuroprotection against A ß1-42 (ß-amyloid peptide 1-42), alternative therapeutic agent for dementia-related neurological conditions, including Alzheimer's disease.	26, 27
6.	D- (+)-Carvone	Antioxidant, antinociceptive, anti-cancer, insecticidal, blood lipid-lowering activity,calcium-channel blocker, antisprouting agent during tuber storage, bactericidal and fungicidal activities.	26, 28, 29
7.	L-Perillaldehyde	Antioxidant, antibacterial, antimicrobial,anticancer, anti-allergic, anti-inflammatory, neuroprotection activity, antidepressant.	30, 31
8.	Bornyl acetate	Antimicrobial, antibacterial.	32
9.	Cyclosativene	Cytotoxic, genotoxic, antioxidant, anticarcinogenic.	33, 34
10.	Geranyl acetate	Antifungal, anti-inflammatory properties	35
11.	Sativene	Antiplasmodial activity, cytotoxicity.	36
12.	Caryophyllene oxide	Non-toxic, non-sensitizing, antifungal,insecticidal/antifeedant, anticancer properties, antimicrobial, antioxidant, antibacterial, genoprotective, anti-inflammatory,chemosensitizing, antiproliferative, analgesic.	37, 38
13.	Epiglobulol	Antibacterial, uterus relaxant.	39
16.	(-)-Caryophyllene-(II)	Analgesic, anti-inflammatory, anti-oxidant, neuroprotective, antimalarial, antiproliferative, anti-convulsant, antifungal, antispasmodic, anti-allergic, anxiolytic/antidepressant, hypoglycemic, local anaesthetic, hypolipidemic, genoprotective, chemosensitizing.	38
17.	.delta-Neoclovene	Antibacterial, antitumor, cardio-vascularprotection	40
18.	Longiverbenone	Antibacterial, cytotoxic activity	41
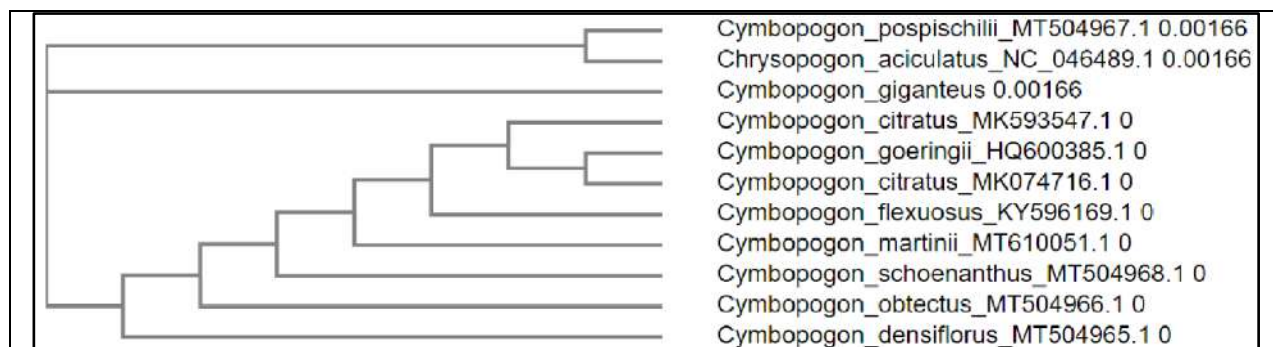
Fig 1: Phylogenetic Tree Constructed Based on rbcl Gene Nucleotide Sequences Of Cymbopogon Species
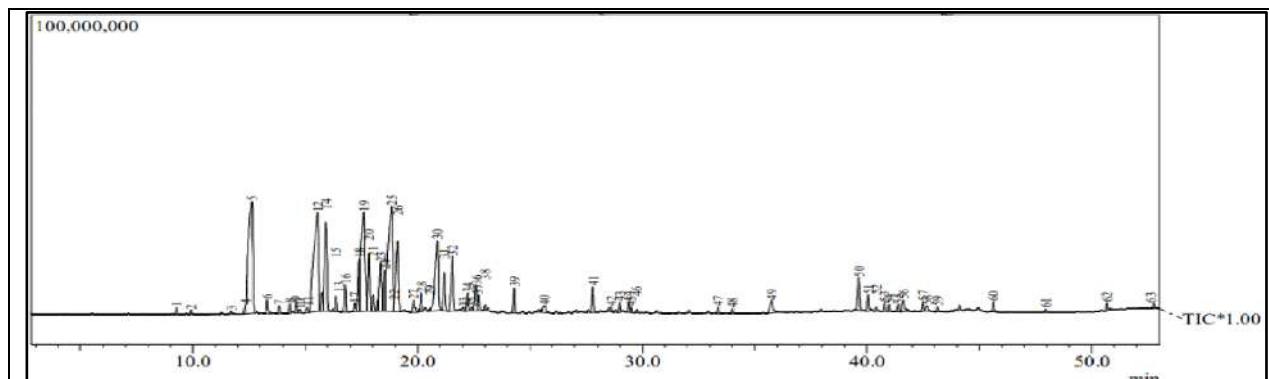
Fig 2: GC-MS Chromatogram of Wild *Cymbopogon Giganteus*
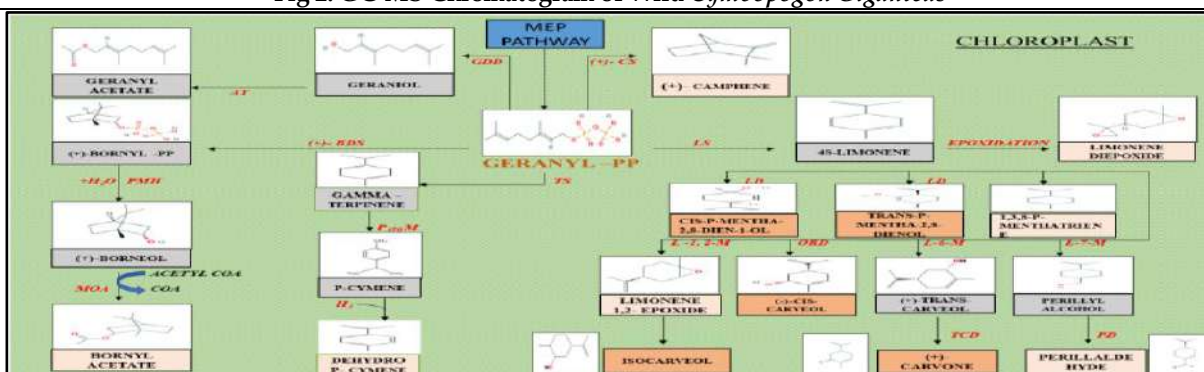
Fig 3: Overview of Monoterpenoid Biosynthesis Pathway in wild *Cymbopogon giganteus*.

Monoterpenoidsare synthesized by MEP pathway in chloroplast from Geranyl Diphosphate as their precursor where orange colour represents the key compounds found in the wild *C. giganteus*, pink colour for compounds reported in the wild *C. giganteus* and grey colour is the intermediate compounds present in the pathway. The enzymes identified via KEGG and META CYC include: **GDD** Geranyl- diphosphate diphosphatase, **AT** Alcohol O- acyltransferase, **(+)- CS** Camphene synthase, **(+)- BDS** (+)- Bornyl diphosphate synthase, **PMH** Phosphoric monoester hydrolases, **MOA** Monoterpenol O- acetyl transferase, **TS** Gamma- terpinene synthase, **P450M** Cytochrome P_{450} monooxygenases, **LS** (4S)- Limonene synthase, **LD** Limonene dehydrogenase, **L-1,2-M** Limonene- 1,2- monooxygenase, **ORD** Oxidoreductase, **L-6-M** Limonene- 6- monooxygenase, **L-7-M** Limonene- 7- monooxygenase, **TCD** (+)- trans- carveol dehydrogenase and **PD** Perillyl- alcohol dehydrogenase.
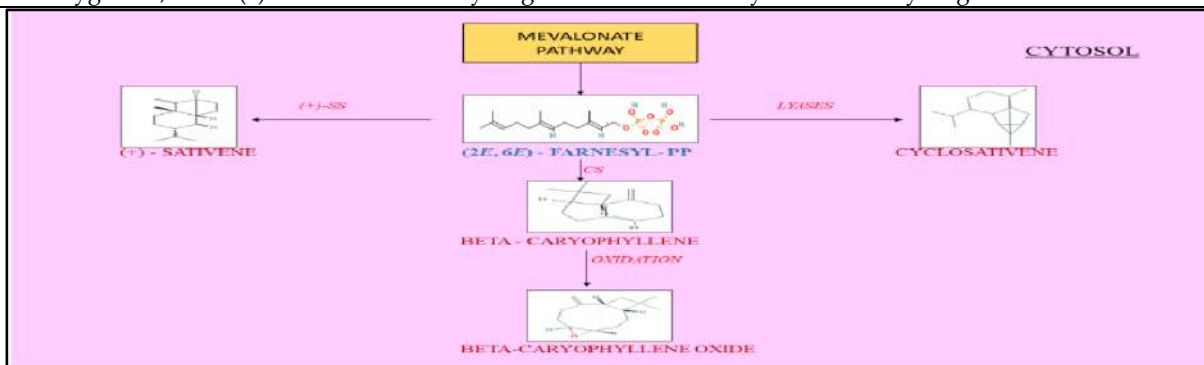
Fig 4: Overview of Sesquiterpenoid Biosynthesis Pathway in wild *Cymbopogon giganteus*

Sesquiterpenoids are synthesized by MVA pathway in cytosol from Farnesyl Diphosphate as their precursor. The enzymes identified via METACYC include: **(+)- SS** (+)- Sativene synthase and **CS** (-)- beta caryophyllene synthase.

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RESEARCH ARTICLE

Protein Functional Annotation and Network Analysis in Relation to Oral Pathogen and Oral Cancer using Computational Tools

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ABSTRACT

Oral cancer is becoming more common, and it threatens to be a serious worldwide medical issue. Approximately 100 trillion microorganisms' cells live in harmony with their host in the human body. Bacteria have long been thought to have a role in immunological regulation, disease onset, and maintenance of optimal health at certain body locations. This study aims to elucidate the networks between proteins and biologically active compounds, as well as their functional annotations, cell signaling pathways. According to the results, online STRING software was used to create a molecular genetic interaction network azurin on oral bacterial proteins, and Cytoscape software was used to calculate 11 nodes, 16 edges, and an average node order of 2.91 and average value. The local clustering factor is 0.876, the expected number of edges is 10, and the enrichment ppi has a p-value of 0.0553. Nodes, lines and colors prove the rationality of interactive networksFor interleukin 8 in oral squamous cell carcinoma, a molecular genetic interaction network is obtained and Cytoscape software is used to visualize the number of nodes 11, the number of edges 55, the average node order 10, and the average. Did. The local clustering factor is 1, the expected number of edges is 20, and the enrichment p-value is 3.25e11This is the only experimentally determined bacterial azulin protein, rubA2, rubA1, oprC, nirS, nirS, nirM, exaB, exaB, ccoQ2, ccoQ1, rubA1, azu, nirM, exaB. Responsible for annotated interactions in the STRING database. For oral cancer proteins, interactive network proteins such as CCL2, CCL3, CSF2, IL10, IL1A, IL1B, IL6, TNF, IL10 according to the gold standard set data.

Keywords: Networking, STRING, Oral cancer, Bacteria

INTRODUCTION

Head and neck tumors are one of the 10 most common types of cancer in the world [1]. Oral squamous cell carcinoma (OSCC) in men is the most common cancer of all head and neck squamous cell carcinomas (HNSCC). It accounts for about 90% of oral malignancies and accounts for more than 300,000 newly diagnosed cases each year [2]. The number of HNSCC cases is estimated to increase to 1 million by 2030 (http://www.who.int/entity/healthinfo/statistics/bod_dalybywhoregion.xls?ua=1). Despite significant advances in cancer treatment and management, OSCC-related mortality rates have not changed. The molecular basis for aggressive OSCC growth and metastasis is still unknown. OSCCs often remain undetected at the high stages associated with high mortality and are therefore associated with high personal and social costs. The 5-year overall survival rate is estimated to be about 50% [3].Therefore, reducing oral cancer mortality requires new targets for early detection and treatment of OSCC. In recent years, identification of genes associated with complex diseases has become an important issue. Experimental approaches, such as B. genetic linkage association studies [4], expression profiling [5], and genome-wide association studies [6], are for genes at high relative risk for diseases such as cancer [7] and asthma [8]. It has been shown to be successful in identifying, Diabetes [9] and hypertension [10]. However, disease heterogeneity, the complexity of finding genes at specific loci, and the associated costs have led to the development of various in silico approaches to the identification of diseased genes.The correlation between a particular bacterium and various diseases, including cancer, is well known and well known. The role of bacteria in some types of cancer has been elucidated in more recent studies, but remains unclear in many other studies [11]. Assessing the dynamics of a microbial population under a medical condition helps determine the mechanisms by which bacteria can be used to induce or develop cancer. Periodontal disease is caused by a bacterial infection of the periodontal tissue that causes gingival inflammation and periodontal disease. Periodontitis affects the gingival tissue but not the underlying tooth support structure. Periodontal disease, in contrast, is an inflammatory disease that extends deep into the tissue and results in the loss of supporting connective tissue. Periodontitis can lead to loss of connective tissue and bone support and is the leading cause of tooth loss in adults [12]. Caries is the destruction of tooth structure by oral bacterial acids produced by microbial fermentation of leftover food [13]. Three characteristics of bacterial species are involved in biofilm formation, acid production, and caries development, including acid resistance [14] Marsh, 2005). Many acid-forming and acid-forming bacteria are involved in the carrier, including Streptococcus mutans and *Streptococcus sorbinus* (collectively known as mutans *Streptococcus*), as well as other acidic strains of lactobacillus and non-mutans streptococcus. [15]. This study follows an observational study design aimed at screening for virulence factors in the oral microflora with these proteins or oral cancer and chewing tobacco complex pathogens. This can probably interact with certain proteins. The interaction of oral cancer protein cross-linking with oral mediator proteins and other proteins was analyzed using the STRING v.5 pipeline [16] (Szklarcz). These nodes were included in the STRING database and the queries were user-defined.

MATERIAL AND METHODS

Sources and Selection of the targeting protein
The source of the plant, the geographic location of the collection, the chemical structure, and the biological activity of theoral bacterial protein azurin and oral cancer cyclin B1 protein were obtained from bibliographic sources, The origin of the plant, the geographic location of the collection, the chemical structure and biological activity of the oral bacterial proteins azurin and oral cancer cyclin B1 proteins were obtained from bibliographic sources including major journals, masters and papers, chapters of natural product chemistry. Unpublished textbooks and meeting minutes. The following standards are used by Kumar et al. [17] and Gupta et al. [18]

Protein network interaction
Extensive network analysis is used to identify functional connections between proteins and emphasize the biological importance of genes linked to enrichment pathways. The STRING and STITCH (version: 11.0) databases are global resources for predicting functional connections between proteins and cloud cluster networks and are used to study

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the interactions between proteins encoded by specific genes. Masu [19]. Experiments to identify species using P-proteins and chemicals as input gene sets. Study protein interactions (PPIs) between enriched genes and networks of interactions between them. A full score above 0.7 indicates a high level of confidence in the presence of significant interactions

RESULTS AND DISCUSSION

Protein-protein interactions are a central part of the cell network and are known to have many effects. Analyze the information flow network between all target proteins to determine the amount of information that flows between cytochrome proteins and other proteins. Use online STRING software to create a molecular genetic interaction network azurin on oral bacterial proteins, and use Cytoscape software to get 11 nodes, 16 edges, an average node order of 2.91, and visualization averages. I calculated in Table 2. The local clustering factor is 0.876, the expected number of edges is 10, and the enrichment ppi has a p-value of 0.0553. Nodes, lines, and colors show the rationality of interactive networks as shown in Figure 1 and Table 1. For interleukin 8 in oral squamous cell carcinoma, a molecular genetic interaction network was obtained and the software Cytoscape was used to visualize the number of nodes 11, the number of edges 55, the average node order 10, and the values. It was made into. The local clustering factor is 1, the expected number of edges is 20, and the enrichment p-value is ppi 3.25e11 as shown in Figure 2 and Table 3.Nodes, lines, and colors prove the rationality of interactive networks and annotation with different genes Table 4. Many experiments have shown that the gene is associated with protein expression. The results show that more residues than the cytochrome-protein cross-linking pathway are involved in protein signal intensity.

The result is represented by the color displayed in the forecast tree. A random machine learning approach [20] was used to build a reliable PPI network by combining co-movement scores obtained from metabolic partitions with detailed information suggesting physical links (Figure 2A).This is because physically interacting proteins exert similar biological activity, co-express, and often have similar evolutionary conservation. Only protein pairs with solid biochemical evidence (at least 0.4 correlation rating) from the fractionated dataset were used, and other supporting features were implemented in this subset. Two additional measurements were recorded from biochemical fractional records that reflect reproducibility. Specifically, there are a number of fractionation experiments in which the co-migration assessment of the protein pair is at least 0.4, and a number of fractionation experiments with the largest peak in each migration profile. Duplicate protein pairs. Other evidence supporting functional associations is co-[deletion] [21,22,23], interacting domains [24,25], co-evolution [26] and phenotypic records [27] (Table 3).Machine learning classifiers trained on a gold standard set containing only experimentally determined bacterial azulin proteins rubA2, rubA1, oprC, nirS, nirS, nirM, exaB, exaB, ccoQ2, ccoQ1, rubA1, azu, nirM, exaB and tested. (Table 2) is responsible for the annotated interactions in the STRING database [18]. For oral cancer proteins, interactive network proteins such as CCL2, CCL3, CSF2, IL10, IL1A, IL1B, IL6, TNF, IL10. Assessment of the relative contribution of each property to PPI prediction (measured by GiniScore) confirmed that the combined biochemical evidence had the greatest effect on classification (Table 4) and was associated with other functional associations. Compared to better co-mobility, it shows that it reflects information and is used to predict interactions.

CONCLUSION

Azurin, putative bacteriocins that have useful properties like those of azurin have been distinguished in more microscopic organisms' species. The systems of azurin and the azurin-like bacteriocins will give more and better choices in oral malignant growth treatment. In this article, we sum up how azurin and the determined peptides capture key cell controllers or cell surface receptors to redesign the phone flagging organizations. Interleukin 8 cytokines might add to the pathogenesis of this sickness, is a significant chemokine of interest in periodontal infections. The concentrate likewise reasoned that the collaboration of protein networks with different proteins can be utilized as potential remedial medication possibility for the avoidance of oral sickness.

CONFLICT OF INTEREST

None declared

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Table 1: Enrichment pathways and linked genes and nodes

Parameter	Value
Number of nodes	11
Number of edges	16
Average node degree	2.91
Avg. local clustering coefficient	0.876
Expected number of edges	10
PPI enrichment p-value	0.0553

Table 2: Networking of Azurin in oral bacterial proteinwith Annotation

Node1	Node2	Annotation node 2	Annotation node 1	Scoring function
RubA2	RubA1	Involved in the hydrocarbon hydroxylating system	which transfers electrons from NADH to rubredoxin reductase and then through rubredoxin to alkane 1 monooxygenase	0.498
RubA2	Azu	Azu transfers electrons from NADH to rubredoxin reductase and then through rubredoxin to alkane 1 monooxygenase	Transfers electrons from cytochrome c551 to cytochrome oxidase	0.685
RubA1	RubA2	Involved in the hydrocarbon hydroxylating system	It transfers electrons from NADH to rubredoxin reductase and then through rubredoxin to alkane 1 monooxygenase	0.498
RubA1	Azu	It transfers electrons from NADH to rubredoxin reductase and then through rubredoxin to alkane 1 monooxygenase	Transfers electrons from cytochrome c551 to cytochrome oxidase	0.680
OprC	Azu	Putative copper transport outer membrane porin OprC; tonB-copper: TonB-dependent copper receptor	Transfers electrons from cytochrome c551 to cytochrome oxidase	0.762
NirS	NirM	Annotation not available	Electron donor for cytochrome cd1 in nitrite and nitrate respiration	0.984
NirS	Azu	Annotation not available	Transfers electrons from cytochrome c551 to cytochrome oxidase	0.855
NirM	NirS	Electron donor for cytochrome cd1 in nitrite and nitrate respiration	annotation not available	0.984

NirM	ExaB	Electron donor for cytochrome cd1 in nitrite and nitrate respiration	Is an essential component of the ethanol oxidation system that allows P.aeruginosa to grow on ethanol as the sole carbon and energy source.	0.691
NirM	CcoQ2	Electron donor for cytochrome cd1 in nitrite and nitrate respiration	Annotation not available	0.874
NirM	CcoQ1	Electron donor for cytochrome cd1 in nitrite and nitrate respiration	Annotation not available	0.853
NirM	Azu	Electron donor for cytochrome cd1 in nitrite and nitrate respiration	Transfers electrons from cytochrome c551 to cytochrome oxidase	0.968
ExaB	NirM	Is the direct contact between electron acceptor of the quinoprotein ethanol dehydrogenase (QEDH)	Electron donor for cytochrome cd1 in nitrite and nitrate respiration	0.691
ExaB	Azu	the direct electron acceptor of the quinoprotein ethanol dehydrogenase (QEDH)	Transfers electrons from cytochrome c551 to cytochrome oxidase	0.692
CcoQ2	NirM	Annotation not available	Electron donor for cytochrome cd1 in nitrite and nitrate respiration	0.874
CcoQ2	CcoQ1	Annotation not available	annotation not available	0.872
CcoQ2	Azu	Annotation not available	Transfers electrons from cytochrome c551 to cytochrome oxidase	0.741
CcoQ1	NirM	Annotation not available	Electron donor for cytochrome cd1 in nitrite and nitrate respiration	0.853
CcoQ1	CcoQ2	Annotation not available	annotation not available	0.872
CcoQ1	Azu	Annotation not available	Transfers electrons from cytochrome c551 to cytochrome oxidase	0.737

Table 3: Enrichment pathways and linked genes and nodes

Parameter	Value
Number of nodes	11
Number of edges	55
Average node degree	10
Avg. local clustering coefficient	1
Expected number of edges	20
PPI enrichment p-value	3.25e-11

Table 4: Networking of interleukin 8 in oral squamous cell carcinoma with Annotation

Node1	Node2	Node1 annotation	Node2 annotation	Score
CCL2	CCL3	May be involved in the recruitment of monocytes into the arterial wall during the disease process of atherosclerosis; Belongs to the intercrine beta (chemokine CC) family	Binds to CCR1, CCR4 and CCR5. One of the major HIV-suppressive factors produced by CD8+ T-cells. Recombinant MIP-1-alpha induces a dose-dependent inhibition of different strains of HIV-1, HIV-2, and simian immunodeficiency virus (SIV); Belongs to the intercrine beta (chemokine CC) family	0.921
CCL2	CSF2	Chemotactic factor that attracts monocytes and basophils but not neutrophils or eosinophils. Augments	Granulocyte-macrophage colony-stimulating factor; Cytokine that stimulates the growth and differentiation of	0.988

		monocyte anti-tumor activity. Belongs to the intercrine beta (chemokine CC) family	hematopoietic precursor cells from various lineages, including granulocytes, macrophages, eosinophils and erythrocytes; Belongs to the GM-CSF family	
CCL2	CSF3	C-C motif chemokine 2; Chemotactic factor that attracts monocytes and basophils but not neutrophils or eosinophils. Augments monocyte anti-tumor activity.	Granulocyte colony-stimulating factor; Granulocyte/macrophage colony-stimulating factors are cytokines that act in hematopoiesis by controlling the production, differentiation, and function of 2 related white cell populations of the blood, the granulocytes and the monocytes-macrophages. This CSF induces granulocytes; Belongs to the IL-6 superfamily	0.984
CCL2	CXCL2	Belongs to the intercrine beta (chemokine CC) family	Hematoregulatory chemokine, which, in vitro, suppresses hematopoietic progenitor cell proliferation. GRO-beta(5-73) shows a highly enhanced hematopoietic activity	0.984
CCL2	CXCR2	Has been implicated in the pathogenesis of diseases characterized by monocytic infiltrates, like psoriasis, rheumatoid arthritis or atherosclerosis. Belongs to the intercrine beta (chemokine CC) family	C-X-C motif chemokine 2; Produced by activated monocytes and neutrophils and expressed at sites of inflammation. Binds to IL-8 with high affinity. Also binds with high affinity to CXCL3, GRO/MGSA and NAP-2	0.929
CCL2	IL10	C-C motif chemokine 2; Chemotactic factor that attracts monocytes and basophils but not neutrophils or eosinophils. Augments May be involved in the recruitment of monocytes into the arterial wall during the disease process of atherosclerosis; Belongs to the intercrine beta (chemokine CC) family	Interleukin-10; Inhibits the synthesis of a number of cytokines, including IFN-gamma, IL-2, IL-3, TNF and GM-CSF produced by activated macrophages and by helper T-cells; Belongs to the IL-10 family	0.989
CCL2	IL1A	Augments monocyte anti-tumor activity. Has been implicated in the pathogenesis of diseases characterized by monocytic infiltrates, like psoriasis, rheumatoid arthritis or atherosclerosis.	IL-1 proteins are involved in the inflammatory response, being identified as endogenous pyrogens, and are reported to stimulate the release of prostaglandin and collagenase from synovial cells	0.983
CCL2	IL1B	Has been implicated in the pathogenesis of diseases characterized by monocytic infiltrates, like psoriasis, rheumatoid arthritis or atherosclerosis.	Promotes Th17 differentiation of T-cells	0.993
CCL2	IL6	Chemotactic factor that attracts monocytes and basophils but not neutrophils or eosinophils.	Plays an essential role in the final differentiation of B-cells into Ig- secreting cells Involved in lymphocyte and monocyte	0.994

			differentiation. Acts on B-cells, T-cells, hepatocytes, hematopoietic progenitor cells and cells of the CNS.	
CCL2	TNF	Augments monocyte anti-tumor activity.	it can stimulate cell proliferation and induce cell differentiation. Impairs regulatory T-cells (Treg) function in individuals with rheumatoid arthritis via FOXP3 dephosphorylation. Upregulates the expression of protein phosphatase 1 (PP1), which de [...]	0.992
CCL3	CCL2	Monokine with inflammatory and chemokinetic properties. Binds to CCR1, CCR4 and CCR5. One of the major HIV-suppressive factors produced by CD8+ T-cells. Recombinant MIP-1-alpha induces a dose-dependent inhibition of different strains of HIV-1, HIV-2, and simian immunodeficiency virus (SIV); Belongs to the intercrine beta (chemokine CC) family	Tumor necrosis factor; Cytokine that binds to TNFRSF1A/TNFR1 and TNFRSF1B/TNFBR. It is mainly secreted by macrophages and can induce cell death of certain tumor cell lines. It is potent pyrogen causing fever by direct action or by stimulation of interleukin-1 secretion and is implicated in the induction of cachexia, Under certain conditions	0.921
CCL3	CSF2	C-C motif chemokine 3; Monokine with inflammatory and chemokinetic properties. Binds to CCR1, CCR4 and CCR5. One of the major HIV-suppressive factors produced by CD8+ T-cells. Recombinant MIP-1-alpha induces a dose-dependent inhibition of different strains of HIV-1, HIV-2, and simian immunodeficiency virus (SIV); Belongs to the intercrine beta (chemokine CC) family	Granulocyte-macrophage colony-stimulating factor; Cytokine that stimulates the growth and differentiation of hematopoietic precursor cells from various lineages, including granulocytes, macrophages, eosinophils and erythrocytes; Belongs to the GM-CSF family	0.988
CCL3	CSF3	Monokine with inflammatory and chemokinetic properties. Binds to CCR1, CCR4 and CCR5. One of the major HIV-suppressive factors produced by CD8+ T-cells. Recombinant MIP-1-alpha induces a dose-dependent inhibition of different strains of HIV-1, HIV-2, and simian immunodeficiency virus (SIV); Belongs to the intercrine beta (chemokine CC) family	Granulocyte colony-stimulating factor; Granulocyte/macrophage colony-stimulating factors are cytokines that act in hematopoiesis by controlling the production, differentiation, and function of 2 related white cell populations of the blood, the granulocytes and the monocytes-macrophages. This CSF induces granulocytes; Belongs to the IL-6 superfamily	0.986
CCL3	CXCL2	C-C motif chemokine 3; Monokine with inflammatory and chemokinetic properties. Binds to CCR1, CCR4 and CCR5. One of the major HIV-suppressive factors produced by	C-X-C motif chemokine 2; Produced by activated monocytes and neutrophils and expressed at sites of inflammation. Hematoregulatory chemokine, which, in vitro, suppresses hematopoietic progenitor	0.983

		CD8+ T-cells. Recombinant MIP-1-alpha induces a dose-dependent inhibition of different strains of HIV-1, HIV-2, and simian immunodeficiency virus (SIV); Belongs to the intercrine beta (chemokine CC) family	cell proliferation. GRO-beta(5-73) shows a highly enhanced hematopoietic activity	
CCL3	CXCR2	Binds to CCR1, CCR4 and CCR5. One of the major HIV-suppressive factors produced by CD8+ T-cells. Recombinant MIP-1-alpha induces a dose-dependent inhibition of different strains of HIV-1, HIV-2, and simian immunodeficiency virus (SIV); Belongs to the intercrine beta (chemokine CC) family	C-X-C chemokine receptor type 2; Receptor for interleukin-8 which is a powerful neutrophil chemotactic factor. Binding of IL-8 to the receptor causes activation of neutrophils. This response is mediated via a G-protein that activates a phosphatidylinositol-calcium second messenger system. Binds to IL-8 with high affinity. Also binds with high affinity to CXCL3, GRO/MGSA and NAP-2	0.899
CCL3	IL10	C-C motif chemokine 3; Monokine with inflammatory and chemokinetic properties. Binds to CCR1, CCR4 and CCR5. One of the major HIV-suppressive factors produced by CD8+ T-cells. Recombinant MIP-1-alpha induces a dose-dependent inhibition of different strains of HIV-1, HIV-2, and simian immunodeficiency virus (SIV); Belongs to the intercrine beta (chemokine CC) family	Interleukin-10; Inhibits the synthesis of a number of cytokines, including IFN-gamma, IL-2, IL-3, TNF and GM-CSF produced by activated macrophages and by helper T-cells; Belongs to the IL-10 family	0.988
CCL3	IL1A	One of the major HIV-suppressive factors produced by CD8+ T-cells.	Interleukin-1 alpha; Produced by activated macrophages, IL-1 stimulates thymocyte proliferation by inducing IL-2 release, B-cell maturation and proliferation, and fibroblast growth factor activity. IL-1 proteins are involved in the inflammatory response, being identified as endogenous pyrogens, and are reported to stimulate the release of prostaglandin and collagenase from synovial cells	0.984
CCL3	IL1B	Recombinant MIP-1-alpha induces a dose-dependent inhibition of different strains of HIV-1, HIV-2, and simian immunodeficiency virus (SIV); Belongs to the intercrine beta (chemokine CC) family	Interleukin-1 beta; Potent proinflammatory cytokine. Initially discovered as the major endogenous pyrogen, induces prostaglandin synthesis, neutrophil influx and activation, T-cell activation and cytokine production, B-cell activation and antibody production, and fibroblast proliferation and collagen production. Promotes Th17 differentiation of T-cells	0.996
CCL3	IL6	C-C motif chemokine 3; Monokine	It is a potent inducer of the acute phase	0.989

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		with inflammatory and chemokinetic properties. Binds to CCR1, CCR4 and CCR5. One of the major HIV-suppressive factors produced by CD8+ T-cells. Recombinant MIP-1-alpha induces a dose-dependent inhibition of different strains of HIV-1, HIV-2, and simian immunodeficiency virus (SIV); Belongs to the intercrine beta (chemokine CC) family	response. Plays an essential role in the final differentiation of B-cells into Ig- secreting cells Involved in lymphocyte and monocyte differentiation. Acts on B-cells, T-cells, hepatocytes, hematopoietic progenitor cells and cells of the CNS. Required for the generation of T(H)17 cells. Also acts as a myokine.	
CCL3	TNF	C-C motif chemokine 3; Monokine with inflammatory and chemokinetic properties. Binds to CCR1, CCR4 and CCR5. One of the major HIV-suppressive factors produced by CD8+ T-cells. Recombinant MIP-1-alpha induces a dose-dependent inhibition of different strains of HIV-1, HIV-2, and simian immunodeficiency virus (SIV); Belongs to the intercrine beta (chemokine CC) family	Tumor necrosis factor; Cytokine that binds to TNFRSF1A/TNFR1 and TNFRSF1B/TNFBR.. Impairs regulatory T-cells (Treg) function in individuals with rheumatoid arthritis via FOXP3 dephosphorylation. Upregulates the expression of protein phosphatase 1 (PP1),	0.993

Role of interleukin 8 in oral squamous cell carcinoma

Figure 1.Azurin protein network nodes interaction	**Figure 2.Interleukin 8protein network nodes interaction**

RESEARCH ARTICLE

Analysis on Electric Vehicle Batteries Sizing and Comparison of various Battery Packs

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ABSTRACT

In this emerging world, the population is increasing tremendously, as a consequence the need for transportation also increases, which could be non - hazardous to the environment. For this, the electric vehicle will be an alternative source. Though the electric vehicle has a lot of advantages, choosing a wise battery type is a quite challenging one. In an electric vehicle, battery is the main source, which provides power to the motor to drive the vehicle. Due to the optimistic growth of technology, there are a lot of battery types available in the market. The overall efficiency of the electric vehicle depends on the selection of battery type. In this review paper, all the battery specifications have been compared based on the data available on the data sheet. The battery pack calculation is also done for selecting the desirable battery for an electric vehicle.

Keywords: Electric Vehicles, EV Batteries, Li-ion batteries, Energy density, Power density.

INTRODUCTION

In this emerging world, the population is increasing tremendously where the transportation is the basic need to fulfill everyone's desire. The non- renewable resources are decreasing day by day due to increase in number of people using transportation. So the way of transportation should be eco-friendly and the electric vehicle will be an alternative source [1]. Though electric vehicle has lot of advantages, choosing a wise battery type is quite challenging one in electric vehicle designing. In electric vehicle battery is the source of electric power which stores chemical energy and

converts it into electrical energy [2]. Many types of batteries with different specification are being manufactured and it should be selected according to the range of the vehicle [3-4]. The number of cells for a battery pack should be selected according to the range of vehicle. While choosing the battery voltage, current, C-rate, energy density, power density should be considered [2,4]. The block diagram of pure electric vehicle is shown in the fig. 1. The power from the battery can be used to drive the motor through the controller. The power should be sufficient to drive the motor, controller and other electrical components. The battery in the vehicle can be charged outside and it will take certain time to get charged completely. While selecting battery, both the Battery life and charge cycle should be taken into consideration because fast charging may damage the battery [6-7].

The battery's nomenclature should possess physical dimensions and electrical characteristics of a particular portable drive cell. British national standards, ANSI, JIS have developed the national standards for dry cell [8]. The battery manufacturers should undergo certain international standards. IEC 60086 is one of such international standards which uplift the battery manufacturers to make sure that batteries are replaceable corresponding to the function, standard form and fit [9]. The manufacturers and end users will be benefited by using this kind of batteries. The following are the two main standards, UL 2054 – Household and commercial standards, UL 1642 – Standard for lithium batteries. The standards UL 2054 and UL 1642 are used as the proof by the manufacturers developing medical devices using batteries to ensure the safety and effectiveness of the device.

Battery Parameters

Batteries are the primary source for Electric vehicles. It has many parameters in terms of electrical, chemical and physical means. The following parameters are essential for electric vehicle battery design [10-11]. All the parameters are varied with respect to the manufactures, chemical properties, physical properties, etc. The exact values of the parameters are only available in the datasheet given by the manufactures [12-14].

Cell Voltage

Voltage, also Known as electric potential difference is the Potential difference between two electrodes (cathode, anode) in a cell. 3.2 - 3.7 is the nominal voltage of lithium-ion cell. 4.2 - 4.35 is the required voltage to charge lithium-ion cells. The basic voltage essential for an electric vehicle is 72V, to secure this voltage the cell is connected in series.

Ampere hour

The amount of charge between two electrodes is called electric current. The unit of charge is Ampere hour. The batter's capacity is expressed in Wh, W, Ah. Among this Ah is the most used one. If a battery is capable of delivering one ampere of current in one hour from the available charge in it, it is known as one Ampere hour.

Power Density

Power density which is also called volume power density is the amount of power per unit volume. The power density is said to be the volume of all energy pack. This is expressed as W/m3. The acceleration of an electrical vehicle depends on power density. Generally, batteries have high power density since its rate of charging or discharging of the battery may affect the charging duration.

Energy Density

In the given region the amount of energy stored per unit volume is known as energy density. The unit of electric density is J/m^3. The range of electric vehicle depends on energy density. To obtain high range for an electric vehicle the energy density should be high. Usually batteries have high energy density, irrespective of their size. A battery is said to be efficient if it is capable of storing large amount of energy in smaller volume.

Weight

The total weight of the battery includes the total cells that is connected in parallel and series. Weight is the major while selecting the cells. As the weight of the cells increases the load of the battery increases as a result the range of the vehicle decreases.

CRATE

In a battery the discharge limit of the current is calculated using Crating. The amount of current required for charging and discharging of a battery is indicated by Crate. From rating of ampere hour the Crate can be calculated. The crate of the battery depends on battery capacity. For better performance of battery crate should be high. From Fig.2 the power density is high for nickel metal hydride. On comparing both lithium ferro phosphate and lithium-ion battery, lithium ferro phosphate has low power density than lithium ion battery. From Fig 2, it is concluded that nickel metal hydride has high power density, hence the acceleration will be high when compared to other batteries. From Fig.3 the energy density is high for graphene battery. On comparing both lithium ferro phosphate and lithium ion battery, lithium ferro phosphate has energy density than lithium ion battery. From Fig 3, on comparing lithium ferro phosphate, lithium ion, nickel metal hydride. It is concluded that lithium ferro phosphate has high energy density, hence the range of the vehicle will be high when compared to other batteries. From Fig.4 it is analyzed that the charging time of the graphene battery is low when compared to other batteries. On comparing both lithium ferro phosphate and lithium ion battery, the charging time of lithium ion battery is low. On comparing lithium ferro phosphate, lithium ion, nickel metal hydride. It is concluded that nickel metal hydride has high charging time.

Comparison of Battery Components

To select the wise battery it is necessary to analyse some of the essential parameter like anode, cathode, electrolyte, power, Crate, Power density, Energy density, cell Ah, life cycle, cell name. The above mentioned parameters depend on each other for a battery to perform in an efficient way. The above mentioned parameters depend on each other.

Battery Pack Calculation

Battery is the most significant part for an electric vehicle. Battery pack calculation helps to decide the number of cells present in a battery pack. Friction, aerodynamic drag, grading weight all these factors may influence the output of a battery. Acceleration, braking, climatic conditions at a particular time helps to determine the drive cycle. Drive cycle varies according to the region. The total power delivered from the battery is sum of the power loss due to external factors and the total consumption by the motor. From the total power delivered, the number of cells connected in parallel and series can be calculated. Based on the energy requirement the total capacity of the battery pack is calculated using the data from the datasheet. The mechanical power needed for the vehicle is identified using the parameters such as aerodynamic drag, rolling resistance, grading and weight of the vehicle.

The Electrical power is calculated by means of total tractive power which comprises of all mechanical forces and acceleration or deceleration force. The flowchart for finding the power required is shown in the fig 6. Based on the drive cycle time duration, the energy consumed by the EV at each instant is calculated. Based on the flow chart shown in fig 7, the energy consumption for Electric vehicle is shown in the fig 8. The graph shows the single drive cycle for an EV. The desired range of the vehicle is 60kms, so that the vehicle covers 658 m for a single drive cycle and energy consumed is 12.328 Whr. Hence for 60kms, the energy consumed by the vehicle is 1124.134 Whr. With the help of this energy consumed value, the battery packs can be designed using the flowchart shown in the fig 8. The battery pack is designed for the EV specification given the table 2 based on the flow chart shown in fig 6. The battery cell used for the EV are Panasonic Lithium Ion cell (NCR18650B), Panasonic VRLA Battery cell (LC – PD1217PG), Li- Iron Phosphate (NX ACL9013), Panasonic Nickel Metal Hydride battery cell (HHR370AH). The datasheet values are given in the table 3. The motor voltage is considered as 48V and power of the motor is maximum power required by the vehicle

In this proposed system, a special technique is used that would purify the quality level of the water before it is transferred to the tank where the drinking water is stored. Here, a PH sensor is placed to sense the PH level of the water before the water from municipalities or corporation reaches the water tank. The water's pH level is detected continuously and if the water is more contaminated obviously the pH level is lesser than the neutral pH of drinking water. Under this condition, the pH level is indicated and the flowing of water is suddenly blocked by closing the two valves at the end through the IOT device. Additionally, chloramine gas is stored in the nearest tank. When less pH level is detected, immediately the chloramine which is situated external to the pipe is allowed to flow through the pipe and sprinkled to the water until the pH level of the water gets into the normal level. After the pH of the water becomes

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neutral to the drinking level the pathway of the water differs. There a new passage opens and the water flows to the mineral cartridges and the water is then transferred to the water tanks where it gets stored to drink safely.

CONCLUSION

By utilising the proposed technology, the battery pack and the value of the cells and the discharge time will be calculated accordingly. Moreover, the charging and discharging cycle will be dependent on the discharging and charging of the cells. The cells voltage will be decrease with respect the motor current and the value of output will be affected. The correct power and voltage rating must be selected to get better performance.

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Table 1. Battery Components Comparison

PARAMETERS	LITHIUM ION BATTERY	NICKEL METAL HYDRIDE (HYBRID VEHICLE)	LITHIUM FERRO-PHOSPHATE
ANODE	Intercalated lithium(+ve)	$Ni(OH)_2$	Graphitic carbon electrode with a metallic backing
CATHODE	Graphite (-ve)	Metal Hydride	Lithium iron phosphate ($LiFePO4$)
ELECTROLYTE	LiPF6 (Mostly comprises of lithium salt)	KOH	LiPF6 (Mostly Comprises of lithium salt)
ENERGY DENSITY	676 Wh/L	140-300 Wh/L	325 Wh/L (1200 kJ/L)
POWER DENSITY	243 W/Kg	250-1000 W/Kg	200 W/kg
CELL Ah	3250 mAh	2200 mAh	3200 mAh
LIFE CYCLE	7500 cycles	500 cycles	2000 cycles
CELL NAME	NCR1865B	HHR210A	26650LFP CELL
CRATE	0.12C	0.1C	0.5C

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Table 2 Electric Two Wheeler Specification

Parameters	Value
Weight of the vehicle (including driver)	200
Aerodynamic drag	0.9
Coefficient of rolling resistance	0.013
Speed of the vehicle	As per IDC
Gradient	0o
Range of the vehicle	60Km

Table 3 Cell datasheet

Parameter	Li- Ion Battery cell NCR18650B	VRLA Battery cell LC – PD1217PG	Li- Iron Phosphate NX ACL9013	Nickel Metal Hydride HHR370AH
Length of the cell	0.0653 m	0.15 m	0.0656 m	0.067 m
Diameter/width the cell	0.0185 m (diameter)	0.0746 m (width)	0.0263 m (diameter)	0.0182 m (diameter)
Mass of the cell	0.0485 kg	2.47 kg	0.080 kg	0.060 kg
Capacity of the cell	3.2 Ah	7.2 Ah	2.3 Ah	3.7 Ah
Voltage of the cell	3.6 V	12 V	3.2 V	1.2 V
C-Rate	1	1	0.5C	0.2C
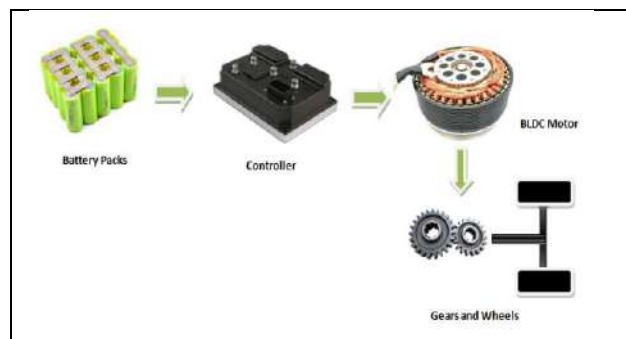
Fig 1. Block Diagram of Pure EV
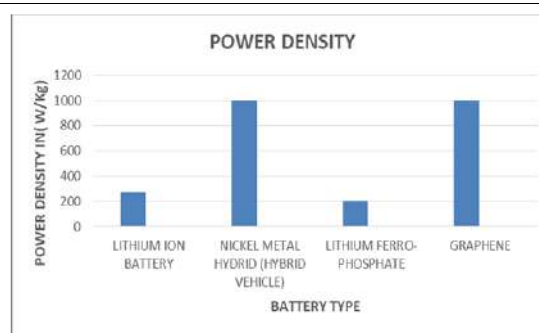
Fig 2. Power density graph
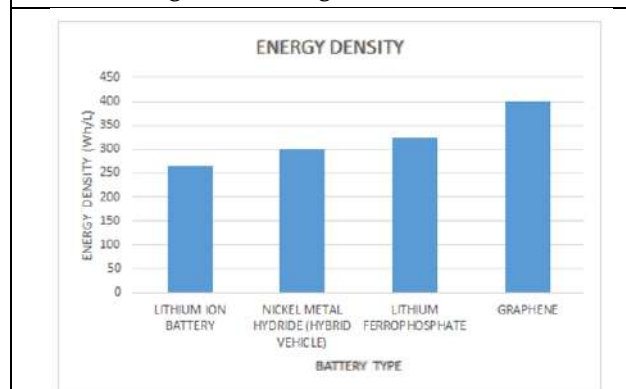
Fig 3. Energy density graph
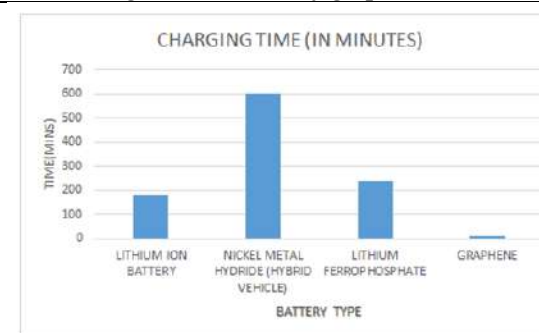
Fig 4. Charging time graph
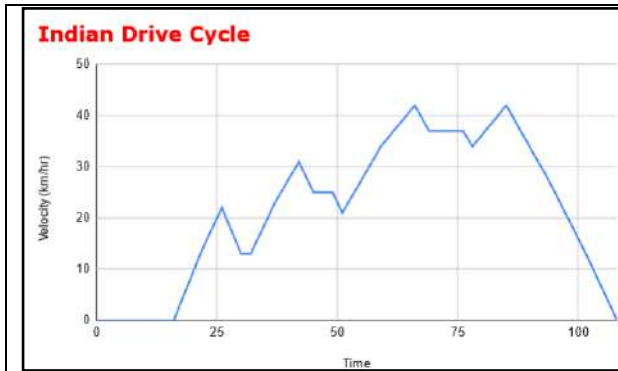
Fig 5. Indian Drive Cycle
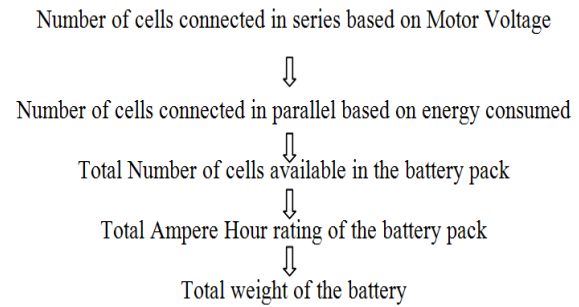
Fig 6. Flowchart for battery pack design
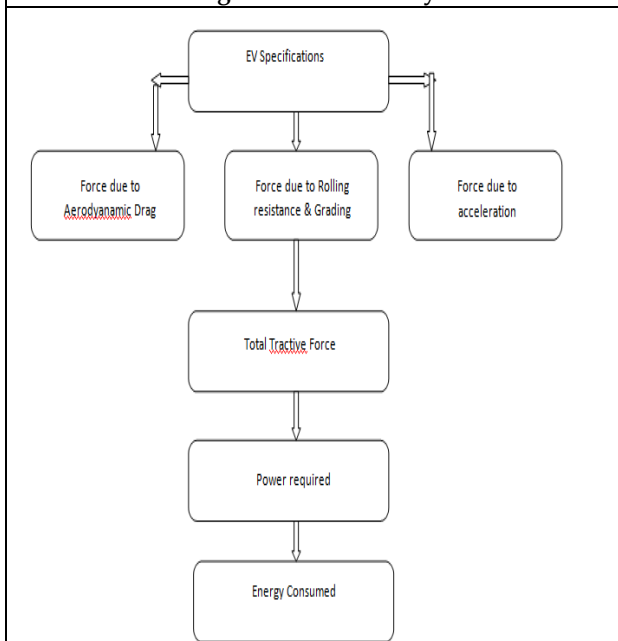
Fig 7. Flow chart for Energy Consumption
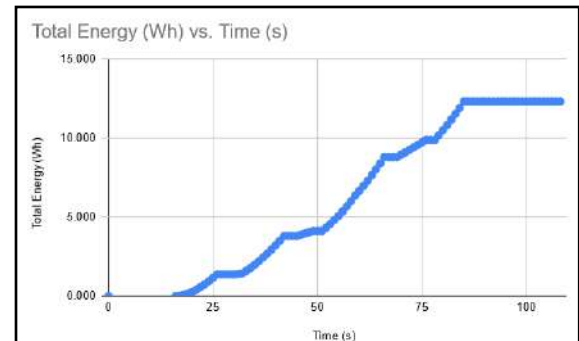
Fig 8. Energy Consumed for IDC

RESEARCH ARTICLE

AROHUB: Educloud Framework for Multicloud Environment

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ABSTRACT

In Contemplory evolution, the multi cloud computing internet based technology which is accessed from anywhere at any time with high speed internet connectivity. The cloud services are managed and owned by the Cloud Service Providers (CSPs). The cloud services used by the clients called are Cloud Service Consumers (CSCs).The users with high speed internet connection can access the cloud service. The Service Level Agreement (SLA) is legal negotiation which is established between the service providers and the consumers. Multi Cloud is an environment with larger numbers of service providers available to serve the requests from the users. Since there are large numbers of cloud users and providers, The Scheduling problem arises in a multi cloud system when a set of user defined tasks is to be mapped to the available set of VMs in all Clouds. ARO_HUB framework is designed to provide the VMs resources to the customers on-demand for minimum time and cost with increased resource utilization. To provide better QoS (Quality of Service) and to meet SLA issues , to satisfy the user needs and increase the profit for cloud service providers. The proposed framework has three parts namely Csp's, Csc and resource broker. The cloud resource broker comprises of three algorithms namely RAMTSA (Rate Aware Meta Task Scheduling Algorithm), The deployment of ARO_HUB framework in educational institution can significantly reduce the expenses of human resources and software licenses. In Future, the research can be extended further by achieving a smarter and optimized task scheduling algorithms with additional parameters for independent and dependent task scheduling in multi cloud environment PAMTSA (Priority Aware Meta Task Scheduling Algorithm) and TCAPSO (Time and Cost Aware Particle Swarm Optimization) algorithm. The proposed ARO_HUB framework is applied for Educloud environment. Educloud is a cloud used for educational purpose and it offers e-learning and other educational services to the

students, teachers and administrative users. The existing Educloud environment is vendor dependent or services depend on single service provider. The combination of Educloud with multi cloud environment will increase the productivity of cloud.

INTRODUCTION

The Cloud Computing is being become popular computing platform which offers on demand computing resources such as CPU, RAM, storage and network usage. Multi cloud is an another evolution which offers different services from various Csps. The resource scheduling in multi cloud is a tedious process. The major conflict is 10 to choose the best service from the pool of CSPs. There is a urgent requirement of a novel framework for the multi cloud. There is no consideration of user tasks in existing cloud environment.   To provide better QoS and to meet SLA issues, there is a requirement for unique framework which can satisfy the user needs and increase the profit for CSPs. This paper is organized as follows The first division gives about the introduction, second division gives about the literature review, third division comprises of framework for Edu cloud. Section 4 has application scenario of Edu cloud and final division deals with the conclusion.

Related Works

Sukhpal singh *et al.,* [Suk, 14] Sukhpal singh et al., [Suk, 14] explained cloud based resource scheduling framework systems. The proposed framework was applied for workload management for the cloud systems. The cloud workload was identified, analyzed and clustered. The clustering mechanism used K-Means clustering algorithm. .Cloud Sim acts as simulation tool for this model In Contemplory evolution, the multi cloud computing internet based technology which is accessed from anywhere at any time with high speed internet connectivity. The cloud services are managed and owned by the Cloud Service Providers (CSPs). The cloud services used by the clients called are Cloud Service Consumers (CSCs).The users with high speed internet connection can access the cloud service. The Service Level Agreement (SLA) is legal negotiation which is established between the service providers and the consumers. Multi Cloud is an environment with larger numbers of service providers available to serve the requests from the users. Since there are large numbers of cloud users and providers, The Scheduling problem arises in a multicloud system when a set of user defined tasks is to be mapped to the available set of VMs in all Clouds. Sonia

Shahzadi *et al.,* [Son, 17] outlined an Educloud framework for the application of cloud technology in the field of education. The frameworks were designed to provide high quality streamed multimedia services to the students with minimum cost and time.   The proposed framework worked well for deploying the e-resources closer to the students and the teachers. The framework supported scalability with high fault tolerance capacity. Open stack was a platform used to deploy cloud applications. Libi Cloud API was used to supply the resources to the user. Open stack and Libi Cloud API were the two different cloud providers used in this approach . This framework enabled the user to experience the multi cloud environment in the Educloud

Rehman *et al.,* [Reh, 14] developed a cloud migration technique of different CSPs based on the user request. The first phase   selects CSps randomly to initiate the service for the first time use. In the second phase, the services were monitored for both allocated service and for other available service. The fitness functions were used as decision variable and it selects the best service provider for the tasks. The users were given freedom to change service from one service provider to another service provider. Abhishek Singh et al., [Abh, 10] discussed about cloud based e-learning solutions to reduces expenses of CSPs The construction costs of the education information system were reduced and the teaching techniques were convenient to the end users.

Paolo Cemim et al., [Pao, 15] suggested education based cloud application It could easily be deployed using standard local computing resources,. This Model has been divided into five main components: Cloud Controller, Node Controller, Centralized Storage, API Control and User Interface. These components were interconnected and they performed operations related to the education such as gathering, retrieving the e-content and staff assisted applications, etc. This proposed architecture had not implemented in the real world multi cloud entity. Abderhim *et al.,* [Abd, 15] described an architectural framework for cloud based e-learning in higher education. The author had analyzed the positive and negative effects of incorporation of cloud applied solutions Jena *et al.,* [Jen, 14] designed a multi objective PSO for the cloud environment. The framework aids to solve multi objective optimization problem for cloud systems. This model has three parts namely cloud user, cloud manager and cloud data center server. Cloud manager acted as the centralized controller where the scheduling algorithm works to allocate the resources in the cloud. Erik et al., [Eri, 11] presented the novel framework to recover   the challenges in the education . The model comprises of three parts namely predictive elasticity, admission control and scheduling of VM. The predictive elasticity migration of the tasks from one VM to another VM within less time and memory space.

Shen *et al.,* [Shen, 14] analyzed an economical service level objective (SLO) based broker architecture for multi cloud environment. The proposed model allocated the VM resources for storage purpose at minimum cost. Xia Min Zheu et al., [Xia, 17] outlined   a novel work to supply the resources to its consumers on-demand. The task scheduling mechanisms were used to provide resources to satisfy the QoS requirements. The framework comprised of three parts namely scheduling management, tasks type and resource characteristics. Scheduling management consisted of SLA, energy consumption details and list of uncertainty tasks. Sukhpal Singh *et al.,* [Sukh, 17] studied a scheduling framework used for execution of the heterogonous and cluster workloads in the clouds. The algorithm the SLA terms   and it was applied in the field of e-commerce.

Aro Hub Framework

The ARO_HUB framework is designed to administer the VMs resources to the CSC on-demand for minimum time and cost with increased resource utilization. The model comprises of three components namely cloud user, cloud resource broker and CSPs. A cloud user is a customer of the cloud applications and places the requests to the server and the resource broker helps to make it .

ARO_HUB framework is designed to provide the VMs resources to the customers on-demand for minimum time and cost with increased resource utilization. To provide better QoS (Quality of Service) and to meet SLA issues , to satisfy the user needs and increase the profit for cloud service providers. The proposed framework has three parts namely Csp's,Csc and resource broker. The cloud resource broker comprises of three algorithms namely RAMTSA (Rate Aware Meta Task Scheduling Algorithm), The deployment of ARO_HUB framework in educational institution can significantly reduce the expenses of human resources and software licenses. In Future, the research can be extended further by achieving a smarter and optimized task scheduling algorithms with additional parameters for independent and dependent   task scheduling in multi cloud environment PAMTSA (Priority Aware Meta Task Scheduling Algorithm)[12] and TCAPSO (Time and Cost Aware Particle Swarm Optimization) algorithm [13]. The proposed ARO_HUB framework is applied for Educloud environment. Educloud is a cloud used for educational purpose and it offers e-learning and other educational services to the students, teachers and administrative users. The existing Educloud environment is vendor dependent or services depend on single service provider. The combination of Educloud with multi cloud environment will increase the productivity of cloud.. The experimental results proved   that the proposed framework outperformed the existing framework in terms of cost, time and energy consumption. The proposed mechanism maps the users QoS requirements of cloud services to the right IaaS service providers that offers the optimal QoS guarantee solution.. Please do not revise any of the current designations.

A cloud resource broker acts as an intermediate manager between CSP and CSC The user requests are delivered to Cloud broker. The broker acts as centralized manager to schedule the user requests to the server. The broker will have information about the VMs 4 available in the cloud server and have the requests from the cloud user. The user requirements are analyzed in the resource broker and the scheduling algorithms are proposed to lessen the overall execution time and cost of the virtual machines in the multi cloud environment. The cloud resource broker comprises of three algorithms namely RAMTSA , PAMTSA (and TCAPSO. The selection of particular rule is shown the Table.1. The Rule 1 is only applicable for the VMs resources at minimum cost. Scheduling phase is used to select the tasks with minimum execution time and allocate the tasks to the VMs with minimum completion time. The rescheduling phase is used to reschedule the tasks from high speed VMs to the low speed VMs. The rule 2 PAMTSA is applied, if the user gives priority to the tasks. The high priority tasks are allocated to high speed VMs with minimum completion time. The rule 3 TCAPSO is an globalized heuristic algorithm which emphasizes more in cost and time reduction.. The PSO acts as enabler to less time, cost and also it increases the CSPs resource utilization. The Additive weighted method is used as fitness function to match the tasks with available VMs in the multi cloud. Chaotic inertia weight enables higher speed of convergence to reach a global optimal solution (CSPs) are the producer of the cloud. The VMs resources are provided as a service to the cloud user on-demand. Every cloud has a manager server which communicates with other cloud server.

Application Scenario Educloud
According According to IBEF (India Brand Equity Foundation) report [Bar, 18], India has become the second largest market for e learning solutions for cloud . In the year 2021, the cloud consumers for the education may increase to 9.6 million. The e-learning applied in the electronic communication technologies to access the educational curriculum materials outside the class room teaching. The e-learning improves the learning ability of the students 4 when it is compared with traditional learning methods. In the traditional learning, there is lack of interaction with the subject experts and also it requires more cost to interact with the subject experts for direct interaction with the subject experts and acquires less cost when it is compared with traditional method. The existing cloud based e-learning platform is dependent on the single provider. Provided by the drop down menu to differentiate the head from the text.

The request analyzer acts as controller for the user requests and is used to identify type of users (i.e. student, staff or faculty . Request Analyzer grants the permission to allocate the resources. It acts as workload manager to manage the user requests and the scheduler. The RAMTSA algorithm is used to allocate the VMs resources to the end users. The faculty, student and admin are the end users of this application. The RAMTSA algorithm is selected when the user gives input as minimum cost. The VMs are provided to the end users to create and view the experiments in (VCL). The RAMTSA algorithm provides VMs to store and compute the resources in the VCL with minimum cost. The students are given permission experiment and attend the tests. The proposed RAMTSA selects the VMs from the pool of service providers and allocates the tasks 1 to the end users. The RAMTSA is only applicable for the content creation, deletion, and for Creation and viewing experiments in VCL for the faculty The Low speed VMs are emphasized in this algorithm. The RAMTSA works for the student tasks such as viewing the e-material, working with experiments in the VCL and for writing and viewing tests. The proposed algorithm costs around 75 $ for 5hours for the VMs. The VMs resource utilization rate of the cloud server is around 85.78%.

The PAMTSA algorithm is used to allocate the VMs resources 1 to the end users with minimum time. The students and faculty are the primary users of this application. The VMs are provided to the primary users are used to interact directly to the experts. The PAMTSA algorithm requires minimum time and maximum cost. The High speed VMs are selected from the pool 2 of cloud service providers. The Students are given provision to listen, record the experts. The expert talks and discussion were stored and in the high speed VMs. The PAMTSA algorithm direct interaction with the subject experts in the Educloud. The proposed algorithm costs around 150 $

for 2 hours for the VMs. The VMs resource utilization rate of the cloud server is around 80.12%/. The TCAPSO algorithm is used to allocate the VMs resources 1 to the end users with minimum time and cost. The students and faculty are the primary users of this application. The algorithm provides VMs for adding and viewing the attendance details of the students. The proposed algorithm costs around 50 $ for 1 hour for the VMs. The optimized results are obtained in this algorithm. The VMs resource 2 utilization rate of the cloud server is around 88.20%. In the educloud environment, there are three different types of cloud users, namely faculty, student and admin. The cloud users are well defined and their usage pattern is well predicted in advance in this scenario.

The faculty user has control over the content and e-learning materials. The faculty creates and modifies the e-learning materials to the users. The faculty requests 1 are sent to the request analyzer. Request Analyzer checks the scheduler free or busy. It maintains the queue to balance the user requests to the scheduler. The scheduler selects the scheduling 3 algorithms based on the requirements given by the faculty. The resources are allocated to the faculty from the pool of service providers. The RAMTSA[14] is more useful for content creation for Virtual computing lab (VCL) and for the uploading the e-contents server. The PAMTSA algorithm is useful for the Uploading attendance details of the students and live interaction with the subject experts. The TCAPSO algorithm is useful for the tasks such as assessment of student performance and for uploading mark details.

A student acts as an participant of the e-learning application. The student requests 1 are sent to the request analyzer. The student requirements, the scheduler selects the algorithm to process the student user requests. The RAMTSA compute the tasks related to the manipulation of the experiments in the VCL. The PAMTSA view the examination results and mark details 1 of the students. The TCAPSO view the subject experts from different university. An Admin is a user who controls overall e-learning applications in the edu cloud environment. The evaluation reports from the teacher and student are controlled by the admin user.

CONCLUSION

The ARO_HUB The ARO_HUB scheduling framework is implemented in the Educloud environment. This consists of RAMTSA, PAMTSA and TCAPSO algorithms. The scheduler chooses an appropriate algorithm based on the user requests . It is applied in e-learning based application for Edu cloud. The ARO_HUB framework is designed to provide resources to the CSC with minimum cost and time. The deployment of ARO_HUB framework in educational institution can significantly reduce the expenses of human resources and software licenses.

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Table 1. Rules of ARO HUB Framework

Rule No	Rules
1	If the (User Requests for Minimum cost) then choose **RAMTSA (Rate Aware Meta Task Scheduling Algorithm)**
2	If the (user demands for time and priority) then choose **PAMTSA (Priority Aware Meta Task Scheduling Algorithm)**
3	If the (user requests resources with minimum time and cost) then choose **TCAPSO(Time and Cost Aware Meta Task Scheduling Algorithm)**
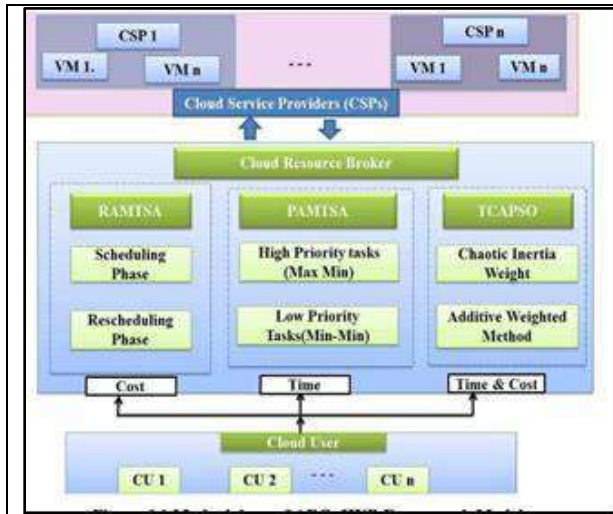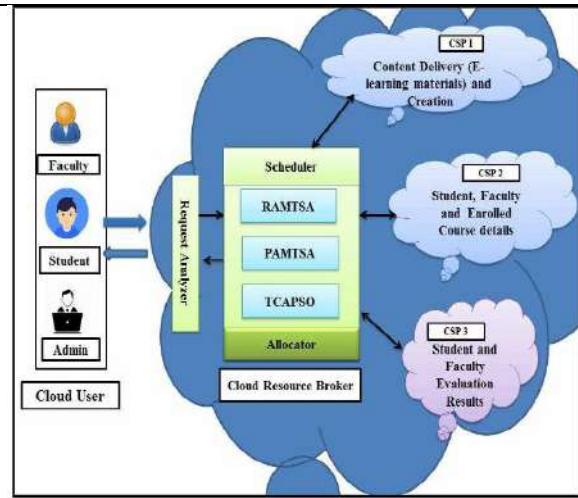
| Fig. 1. ARO_HUB | Fig. 2. ARO_HUB based Edu cloud |

RESEARCH ARTICLE

Corporate Storytelling: Formal Application of Stories in Indian Organizations

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ABSTRACT

The purpose of this paper is to understand the manner in which stories are being used in Indian organisations through phenomenological qualitative research methodology. The paper further aims to understand the issues and the channels of formal communication where storytelling is most utilized. has been adopted for this study. In-depth face to face interviews were conducted with 20 corporate professionals through the Zoom communication platform. From the analysis, 4 themes emerged: why stories (the reasons for storytelling), what stories (the types of stories), whose stories (the managerial positions of the narrators) and how stories (the most effective storytelling mode). It has also been found through analysis that during these trying times, especially with the onset of the pandemic where a lot of norms have been altered, stories that have been shared during meetings and via emails have helped the individuals cope with the new normal. The study will be highly beneficial for the employees and the leaders of corporates as it will enable them to create more effective teams, assign responsibilities more effectively, develop a successful corporate brand and even manage conflicts in a more effective manner. The study will also be beneficial to the students and the teachers as it will enable them to understand the importance of storytelling as an essential skill to be learnt and taught in higher education institutions.

Keywords: Corporate Communication, Storytelling, Formal Communication, Corporate Stories.

INTRODUCTION

It was a hot, summer day. A thirsty crow was circling the skies looking for water to quench its thirst. It was long before it could find a pitcher of water near a tree. It perched itself on the rim of the pitcher and peered inside only to find that the water level in the pitcher was very less making it difficult for it to drink the water. It tried to push its head into the pitcher but the neck was too narrow. It tried to tip the pitcher over to drink the water but failed as the pitcher was too heavy. As it looked around for alternatives, it found some pebbles lying nearby. It dropped one pebble into the pitcher and was ecstatic to find that the water level rose a little. It then started dropping pebbles into the pitcher until the water rose to the level making it easy for the crow to drink the water. It finally drank the water and flew away a happy bird. We have all associated this story with the adage "Where there is a will, there is a way". But, when said in the context of corporate storytelling, the crow could have flown elsewhere when it found that the water in the pitcher was too less for it to drink. But it stayed back, waited, and looked for alternate solutions to the problem at hand thus saving energy, time, and resources. Therefore, when said in the context of corporate storytelling, the story conveys an important message of smart work, perseverance, judicious resource allocation and problem solving.

For this research paper, corporate storytelling is defined as the activity that "designates private and public companies' and organisations' strategic utilization of stories and storytelling (in the broad sense of man's ability to tell and understand narratives) to create coherence and progression concerning the companies' or organisations' brand, identity and development" (Norlyk, Lundhot, and Hansen, 2013). Using this idea as the basis for this study, corporate storytelling is the use of narratives in establishing effective communication with the internal and external stakeholders of the organisation. "Stories" in this research paper include but are not restricted to moral and mythological stories, stories of past personal experiences, stories of success or failure of individuals that have been published in books, newspapers, and the like.

In organisations, stories have proved to have an impact on understanding organisation culture, developing the brand image, communicating corporate strategy, managing and leading change in the organisation, communicating to the external stakeholders (the customers, suppliers, investors, and the public), creating processes and systems and communicating these processes to the teams along with developing and sustaining a diverse workforce (Wilkins, 1984; Herkovitz and Crystal, 2010; Barker and Gower, 2010; Marshall and Adamic, 2010; Ohara and Cherniss, 2010; Gill, 2011; Dolan and Bao, 2012; Gomez- Barris, 2017). The purpose of this phenomenological study is to understand the manner in which stories are used in Indian organisations and to understand the various stories that are used in corporate communication and the communication methods which are most successful in corporate storytelling.

Review of Literature
Table 1: Table Showing Review of Literature

Research Gap
The following research gaps have been found

a) a)Majority of the studies regarding corporate storytelling have been conducted in countries outside India. This study aims to fill this gap by focusing on how storytelling is used by corporate professionals in their day–to–day organisation communication in India.
b) Majority of the studies have looked at corporate storytelling either through literature review or through organisational analysis. This study aims to fill this gap by conducting a qualitative research by interviewing corporate professionals on how storytelling has helped them in organisation communication.
c) There is a gap in understanding the most effective mode of corporate storytelling and in understanding how managerial position determines the impact of the story. An attempt to fill these research gaps has been made through this study.

Research Objectives
The objectives of the study are
1. To understand the prevalence of storytelling in Indian organisations
2. To analyse and categorize the situations where storytelling is used in the organisations in India
3. To understand the forms of formal communication that supplement storytelling in Indian organisations
4. To analyse if the managerial level of the storyteller impacts the effect of the story on the listener

RESEARCH METHODOLOGY

Sample Size
This is a phenomenological study analysing the phenomenon of storytelling in Indian organisations. Based on the guidelines enumerated by Creswell (2013) and Polkinghorne (1989), interviews have to be conducted with 5 to 25 people who have experienced the phenomenon. Accordingly, face-to-face interviews were conducted with 20 corporate employees.

Data Collection
In – depth face – to face interviews using a structured interview schedule, were conducted with 20 corporate professionals who had experienced the phenomenon of storytelling in their organisations. One of the methods of validation of the study as stated by Creswell and Creswell (2018) and Eisner (1991) is Expert Consensual Validation. This validation was obtained from 1 industry expert and 1 academic expert and the suggested changes were incorporated. A participant information form and consent form were sent to the participants to be signed and submitted before the interview. These interviews were conducted online and recorded with the permission of the participants via the Zoom platform owing to COVID – 19. The interviews were scheduled according to the participants' availability and went on from a minimum of 20 minutes to a maximum of 45 minutes. The participants were, therefore, asked structured questions regarding the types of issues where storytelling was used in organisations, the types of stories that were narrated, and the method of narrating the stories (oral or written communication). The recordings of the interviews were then used to transcribe and subsequently code them using the QSR NVivo 10 software. The descriptive data was coded using IBM SPSS software. The themes were then derived and the analysis was made thereafter.

Narrative Paradigm Theory developed by Fisher in 1984 forms the theoretical framework of this study. Narrative here refers to any verbal or non – verbal interpretation that is logically arranged to ensure meaningful communication. The Narrative Paradigm Theory that identifies everyone as storytellers also identifies two aspects of narration: coherence (the degree to which the story makes sense to the listener) and fidelity (credibility and the reliability of the story (Cragan and Shields, 1998; Barker and Gower, 2010).

Data Analysis and Discussion
Descriptive Analysis
Table 1 shows the distribution of the descriptive details of the respondents.

Theme 1 – Why Stories?
This theme analyses the reasons for respondents to use stories in their corporate communication (20 references and 183 references).

Sub-theme 1: Storytelling for Effective Routine Corporate Communication
The first reason for respondents to use stories was for effective routine organisation communication. The sub – theme and the codes have been explained below. The code 'storytelling in everyday organisational communication'(39 files and 18 references) includes all the communication made for the purpose of:

Regulating employee performance

- Ensuring appropriate employee behaviour
- Drawing the attention of the listener with the words "Let me tell you a story".
- Managing the overall development of the organisation and the stakeholders.
- Increasing the relatability to the situation at hand.
- Mentoring and coaching the subordinates.
- "An effective tool of communication" to ensure goal orientation and team work.
- Discussing the developments of a project and improvements to be made.

'Situational storytelling' (11 files and 22 references) included the stories narrated according to the situation at hand because "integrating stories to fit into various situations is the essence of storytelling".

The other codes that were mentioned under this sub-theme included:
Effective inventory management especially in retail businesses (1 file and 1 reference).

- Communicating the project details to the team to make employees understand the reason for the job assignment, their roles and their timelines. (2 files and 3 references)
- New employee orientation along with helping them adjust to the organisation culture, and helping them overcome any initial awkwardness (4 files and 4 references).
- Effective presentations and team meetings while sharing business knowledge with peers and subordinates (2 files and references each).

Sub-theme 2: Storytelling to Build an Effective Organisation Culture

The second reason for respondents to use stories was to build an effective organisation culture (19 files and 49 references). The sub – theme and the codes have been explained below. The code 'helping employees come out of difficult situations' explained the use of narrating stories of past similar situations in helping their counterparts overcome difficult situations by sharing stories of similar situations faced in the past (19 references, 10 files). Using stories of past experiences 'to arrive at more effective solutions for the problem at hand' was another important code (10 files, 11 references). Another respondent explained that supplementing data with stories was effective in solving data related problems as stories helped "draw a picture that connected the dots".

The other codes that were mentioned under this sub-theme include: Coping with uncertainties like the pandemic (5 files and 5 references). In the year 2020, many organisations adapted to this new normal by introducing "story happy hours", "storytelling workshops for the children of the employees" and "starting meetings with stories" to motivate the employees in the changing times. ◉ Creating a safe workspace (4 files and 6 references) by regulating employee conduct and calling out employees for harassment are the common reasons for the use of storytelling in internal corporate communication.

"Enabling collaboration among unlike minds in the organisation", "team meetings" and all other types of team communication that build team bonds (3 files and 3 references).
Process development and orientation (3 files and 5 references) while "conducting feasibility checks", "putting work schedules and processes in place" and "introducing new processes in the organisation" along with communicating those processes to the employees. This also included business analysis and making and communicating process decisions regarding the future growth of the organisation.

Sub-theme 3: Storytelling for Employee Performance Appraisal and Motivation

The third reason for respondents to use stories was to effectively appraise and motivate the employees (15 files, 24 references). The sub – theme and the codes have been explained below.

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With 11 files and 17 references, storytelling was most effective in motivating:

- "teams and the employees when they are overwhelmed with deadlines and projects"
- "employees especially after they lost focus of their purpose in the organisation or
- want to quit"
- "them to give their best, especially during the pandemic situation"
- "the employees after a bad event or a bad quarter".
- "motivating employees to stay back and not leave the organisation"

Performance appraisal and communicating performance feedback in an encouraging manner was another aspect of corporate communication where storytelling was most useful (6 files and 7 references). It is defined as the methods and strategies that the organisation uses to track employee performance and growth and provide subsequent feedback (Dijk and Schodl, 2015). One of the respondents elucidated,

Sub-theme 4: Storytelling for Internal and External Stakeholder Relationships

According to Friedman and Miles (2006) and Cornelissen (2004), stakeholders have been defined as all those individuals, groups, organisations and systems like "employees, customers, shareholders, suppliers, non-government organisations, media etc" that have an interest in the actions of an organisation and can be subsequently affected. The code 'communicating with clients, customers and investors' (20 references,12 files) deals with communication with the external stakeholders. "Product advertisements", "solving customer grievances with stories from past experiences", "explaining the product uses and benefits to the customer" and communicating with investors are the main subjects in corporate communication where storytelling is most advantageous. Another respondent mentioned storytelling as "the bridge" to connect with the customers and clients. Narrating the customer information especially "the kind of customers that we are serving" to the potential investors can make an impact on the investments made by them. Storytelling is also being used for developing employee connect with their peers, seniors, and subordinates (5 files and 14 references). This includes, "bridging the gap between old employees and the new employees." And this aspect according to a few respondents can be achieved through storytelling in meetings or presentations or through organisation newsletters and leader connects. Since "storytelling helps to create a personal bond and break the ice with strangers", fostering bonds and relationships even with potential customers, clients or investors is yet another use of corporate storytelling (3 files and 3 references).

Theme 2 – What Stories?

This theme analyses the types of stories that the respondents use in their corporate communication and is mentioned in 20 files with a total of 71 references.

Sub-theme 1: Personal Experiences

The first type of stories was personal experiences (19 files and 51 references). The respondents explained that sharing 'own personal experiences' that they faced in the past in the current organisation or previous organisations where they worked helped their peers and subordinates to overcome the difficult situations they were facing (17 files and 27 references).

Next, sharing the experiences of other employees including the instances that "other employees including older employees who left the organisation faced in the past" and "success stories of employees in the organisation" were also personal experiences that were shared by the respondents in their regular organisation communication (10 files and 18 references).

"Sharing failure stories were more important than sharing success stories" especially when shared via company newsletters and emails so that the employees would feel motivated in the face of failures (6 files and 6 references). The respondents also stressed that "if the team sees us making those mistakes, they can understand how to do certain tasks better", "it is important to tell the team about what went wrong and how it went wrong as well" and "it is more important to share how the employee can get back after a supposed failure" because "it is important to tell the team that it's okay to fail."

Sub-theme 2: Published Stories
This sub-theme was cited in 7 files and 10 references. This sub-theme explained the type of stories that the respondents quoted from books, newspaper articles and social websites like LinkedIn (including the life stories of famous industrialists and businessmen) in relevant communication to get their point across. Quoting these stories helped to motivate their colleagues to achieve goals as well as personal and professional growth.

Sub-theme 3: Others
This sub-theme included all those stories that were a miscellaneous collection of stories (9 files and 10 references). The respondents explained that citing dialogues including the jokes from movies and recalling scenes from movies were very effective as they helped lighten the atmosphere at a meeting, solve conflicts and communicate the essential message to the team and colleagues (2files and 3 references). The respondents also mentioned that when their leaders shared imaginary stories and examples that were relevant to the situation, it helped them remember the message better and reach their goals easily (2 files and 2 references). Similarly, sharing motivational stories also helped the respondent to communicate with the team and elicit the required results (1 file and 1 reference).

Stories from Indian mythology (2 files and 2 references) were effective mainly because of the possibility of "interconnected stories" that could be used for conveying objectives in big projects. A couple of respondents also mentioned that testimonials and good feedback were types of stories that helped to connect with prospective clients. This finding was also confirmed by Denning (2011) in his book on storytelling. To sum this up one of the respondents explained: … Let's say you're making a sales pitch in your business and when you mention the experience of a client organisation, it is a positive influence.

Theme 3 – Whose Stories?
The theme tries to understand whether the managerial experience and the position of the narrator in the organisation affects the message of the said story (20 files and 30 references). 'Managerial position and experience are more important' elucidated that the respondents believed that seniority affected the impact that the story had on the listener (10 files and 14 references). This was because "a superior has been a part of more happenings not only in the organisation but in the domain as a whole therefore his/her stories are likely to be more credible".

Some respondents also believed that "the story is more important than the managerial experience and position" since "everyone has a story to tell and sometimes the most beautiful stories come from the people who are at grassroot level and can add value to an organisation" (8 files and 12 references).

Theme 4 – How Stories?

This theme analyses the mode of communication in which stories are most effective (20 files and 55 references). Storytelling was more effective in oral communication because "it provided more clarity in sharing experiences especially when major communication is done via emails because expression and voice modulation had the desired impact on the listeners as it enabled visualization especially in all online meetings especially during the pandemic". Moreover "rectifying mistakes became easier through oral storytelling" (18 files and 28 references). "Quotes and short stories in emails helped to and hold the attention of the reader and motivate an employee while giving him directions on how to proceed" (12 files and 19 references). But they also mentioned the importance of short stories in the emails and that separate emails sharing success stories of the employees or employee experience can have the

desired effect. Storytelling through oral means when supplemented with written means made the message more effective (5 files and 8 references).

To sum up
I think it's entirely dependent on the audience, and the person who is delivering the message. Also, it's just about putting all your heart in it because it means something to you. If that's there, I would like to believe that people would listen to you or read what you've written.

DISCUSSION

It can be understood from the analysis that stories in corporate communication are mainly used to build an effective organisation culture; effectively communicate with customers, clients and investors more; to develop employee connect with each other and the organisation; motivating employees via personal experiences especially failure stories; to enable teamwork and collaboration in the organisation; create a successful brand while building and maintaining relationships with internal and external shareholders. These findings were in concurrence with the findings of the studies done by Boje (1991), Sole and Wilson (2002), Kahan (2006), Hermansson and Na (2008), Herskovitz and Crystal (2010), Denning (2011), Gill (2011), Dolan and Bao (2012), Singh and Sonnenburg (2012), Mendonca (2015), Silverman (2006), Slattery (2019) and Anderson and Schill (2019). The analysis in the study also throws light on the importance of narrating stories that are relevant according to the situation at hand. Developing and communicating the processes and helping employees come out of difficult situations are yet other reasons for stories to be used in corporate communication. The study mentions the importance of narrating mythological stories along with sharing the lived experiences (personal or published) of colleagues, coworkers and successful leaders. Some respondents believed that while managerial position and experience was important, the credibility of the stories was equally important. It was also learned from the analysis that sometimes the best stories can come from any corner and the story is more important than the managerial position or experience of the individual. Stories communicated via oral communication had an impact on the listener because of the emotional connect that was created. Written stories could be more effective if they were shorter and embedded into the written means of communication. To sum up the study, storytelling is a means of communication that is undermined in organisations and should be used more for effective corporate communication. It is a skill that should be encouraged in organisations to develop the brand and enable effective communication. It is also a skill that must be taught at the higher education institutions to train the students about effective communication in the workplace.

CONCLUSION

Storytelling is an art and everyone should use it for effective communication. The study enables the understanding of how storytelling is used in corporate communication in organisations in India through 4 main themes. The study expands on the why, what, who and how of storytelling in India. From the analysis, an additional reason for storytelling has emerged. In these trying times especially with the onset of the pandemic where a lot of norms have been altered, stories that have been shared through oral and written means have helped the individuals cope with the new normal. It is therefore important for the managers and employees to acknowledge the importance of stories and use them for effective corporate communication to build a stronger brand image with the internal and external stakeholders. It is also important for the students and the teachers to recognise the importance of storytelling as a skill (for effective communication in various corporate scenarios) that be taught and learnt in higher education institutions.

Limitations of the Study and Scope for Further Study
The study has taken a general view of storytelling in Indian organisations. A sector-specific and an industry-specific study is therefore an essential scope for further study. Also, a study on understanding how storytelling is used in

interdepartmental communication is also an effective scope of study. The study has also taken the responses of the corporate professionals to understand the importance of storytelling in organisations. Research scholars taking up further studies about the importance of storytelling can also take the responses of the students and the teachers to understand the importance given to storytelling as an effective means of communication. The study used the interview technique of data collection. Experimental analyses can also be done to understand how the communication in different departments in the organisation can be affected by the use of stories in the communication.

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Table 1: Table Showing Review of Literature	
Concepts addressed in Literature	**Authors**
Corporate as a holistic understanding of the organisation as one coherent and coordinated body (corpus).	Norlyk, Lundhot, and Hansen (2013)
The essential features of an effective story are: plain, simple, and direct style, credibility, goal and authenticity, congruence between the words and actions, timing and spontaneity of the story delivery, communication channel; sensitivity and connect to the audience	Reissner& Pagan (2013); Denning (2011); Schröder (2015)
The reasons for corporate storytelling are: Sharing customs and beliefs through oral storytelling Problem solving Easy employee transition during organisational change. Sharing of complex tacit knowledge along with enabling the understanding of concepts and strategies Form healthy, diverse and balanced teams Communicate innovative strategies and handle objections Motivating employees. Stopping the spread of rumours Communicating ethical values Brand management and customer brand connect Building rapport with the customers Feedback management Sharing topics that are otherwise taboo	Mitroff and Kilmann (1975); Senge (1990); Boje (1991); Boje (1995); Rosenfield,Giacalone and Riordan (1995); van Riel (1995); Swap, Leonard, Shields and Adams (2001); Sole and Wilson (2002); Abma (2003); Denning (2004); DeLarge (2004); Schultz and Kitchen (2004); Sinclair (2005) Kahan (2006); Silverman (2006); Eck (2006); Adamson, Pine, Steenhoven and Kroupa (2006); Boal and Schultz (2007);Hermansson and Na (2008); Baker and Boyle (2009); Barker and Gower (2010); Herskovitz and Crystal (2010); Marshall and Adamic (2010); Woodside (2010); Denning (2011); Gill (2011); Kowalewski and Waukau-Villagomez (2011); Singh and Sonnenburg (2012); Dolan ad Bao (2012); Anderson and Schill (2013); Lee, Liu, Shi, Khoo and Pang (2013); Spear and Roper (2013); Mendonca (2015); Arnaud, Mills and Legrand (2016); Spear and Roper (2016); D'Arrigo, da Silva, Larentis, Camargo and Schmiedgen (2017); Gherardi, Cozza, Poggio (2018); Chakraborty (2018); Slattery (2019).

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Table 2: Table Showing the Details of the Respondents

Characteristics		Percentage of Respondents
Domain of Experience	IT/ITES	20.0
	Finance	10.0
	Marketing	15.0
	Operations	20.0
	Human Resources	5.0
	Others	30.0
Years of Experience	0-5 Years	5.0
	5-10 Years	40.0
	Above 10 Years	55.0
Managerial Position of the Respondents	Middle Level	45.0
	Top Level	55.0
Respondents' Use of Stories in Formal Corporate Communication	Yes	95.0
	No	5.0

RESEARCH ARTICLE

Comparative Study on Periphytic Algal Flora From Selected Pokkali and Prawn Fields of Pizhala Island

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ABSTRACT

The present study was aimed to investigate the algal influence in the pokkali-prawn fields in the success of pokkali and prawn cultivation. Periphytic algal flora was studied during seasons; post pokkali, middle prawn, and post prawn season, from November 2017 to April 2018.Water quality parameters like temperature, pH, alkalinity, salinity nitrite phosphate silica, and sulfate were estimated and studied in a comparative account. The constant temperature was observed in the first two seasons and increased in post prawn season. Alkalinity increased gradually in three seasons. Nitrite content was higher in the middle prawn season than in the other two seasons. Comparable low phosphate content obtained in post prawn season. Sulfate and silicate concentrations were higher in the last two seasons. Periphytic algal flora from Pizhala island of Ernakulam district was studied during three Pokkali seasons, November-2017 to April -2018. Bacillariophyceae members were dominating during all seasons. The sites were labeled using GPS by Google earth. Water quality parameters like temperature, pH, alkalinity, salinity nitrite phosphate silica, and sulfate were estimated and studied in a comparative account. About 54 species of Bacillariophyceae, 2 genera of each Chlorophyta and cyanophycean were reported.

Keywords: Algal flora, pokkali, GPS, Hydrological parameters and Prawn fields

INTRODUCTION

Pokkali is a rice variety free from chemical fertilizers and pesticides that have a traditional practice of about 3000 years. Pokkali cultivation starts in the month of June and ends in the month of October. After the pokkali harvest prawns are cultivated in the fields which is traditionally called "*ChemmenKettu*". The salinity of water in Pokkali fields reduces in the month of August and is maintained till the end of January, and slowly salinity increases. So, there is a gradual transition from freshwater to saline water in Pokkali fields. A large number of microorganisms

inhabit during this time. The present study is focused to study seasonal variations of algae of Pizhala pokkali farm during different seasons and to study physiochemical water quality in three seasons and also to evaluate whether algae have any ecological significance in the development of prawn.

MATERIALS AND METHODS

Collection Site

Pizhala pokkali ecosystem covers an area of about 90 acres, consisting of about 15 Pokkali fields. Even though Pokkali cultivation is active in all fields, prawn cultivation is restricted to 7 fields and only three sluice gates are present for these 7 fields. The surface area of plot 1 and plot 2 to is 22200 and 27000 sq. meters respectively Two plots which have an individual sluice gate is selected for analysis. Sampling plots were selected in such a way that active Pokkali and Prawn cultivation should go on for at least five years, as well as individual Sluice gates should be there so that the plot has its own boundary.The surface water samples were collected from six sites from a plot during three seasons of Pokkali, post-Pokkali season (14 days after Pokkali harvest, 11/11 /2017), Middle Prawn season (30 days after introducing prawn, 6/2/2018) and post Prawn harvest season (7 days after prawn harvest,18/4/2018).The samples were brought into the lab as the earliest for fresh studies. The qualitative estimation was done by using a compound microscope and photographs were taken using a camera (Nikon E45OO). The photographs obtained were transferred to a computer for identification. Abode photoshop was used for getting the dimensions and platted were carried out using it.The water Physico-chemical parameters like Temperature, pH, alkalinity, salinity, nitrite phosphate, silica,and sulfate were analyzed.The temperature was measured using a thermometer and pH using a pH meter. The titration method was used to estimate alkalinity (Phenolphthalein method) and salinity (Mohr's method). Silica was estimated using IS.3025 (part- 35) and sulfate by IS.3025 (Part - 24).Nitrite was estimated by (SAM-NEDA) and phosphate by the Ascorbic acid method.

RESULTS AND DISCUSSION

Algal Flora – A Comparative Study

The main intention of the present work was to study the algal flora and water-quality variations during different pokkali seasons. Bacillariophyceae members were dominating during all seasons. About 54 species of Bacillariophyceae, 2 genera of Chlorophyta, and cyanophycean were reported Considering factors during identification include cell size, colony, shape, and localization of granules, spines bristles, spikes,etc. were considered. Based on these algae were grouped into taxonomic categories like genus and species.

Prawn-Preference of Algae

"The most preferred algal food by African prawn *Macrobrachium vollenhovenii* was Chlorophyta and Diatoms. *Nitzschia closterium* and *Stephanodi- scus hantzschii* were least preferred. The presence of *Nitzschia sigmoidea* and *N. closterium* was observed in the gut. Euglenophycean members, *Euglena tripteris, E. vorax* and *E. sanguinea* were also seen in the gut analysis" (Fredrick *et al*. 2015). In the present study Chlorophycean members Oedogonium sp. and *Spirogyra* sp. were reported only during the Post Pokkali season, indicating that prawns were checking their population.Diatom species were dominating in all seasons. Genus *Nitzschia* were dominant in all seasons proving the least preference of prawn. *Nitzschia sigmoidea* was reported in post pokkali season and post prawn season, proving the affinity of prawns to that species. Euglenophycean members, *Astasia fustis, Euglena sp*., and *Phacus sp.* were reported during post pokkali season and post prawn season and absent during prawn season stating that prawns might have feed these members. "Cyanophycean members *Oscillatoria sp*., *Spirulina sp.* and Zygophyceaen members *Zygnema sp. Cosmarium sp*. and *Spirogyra sp.* were proven as the natural feed of prawns. Among diatoms *Navicula sp., Cymbella sp*. and *Fragillaria sp.* proved to be the natural feeds of prawns" (Bakhtiyar *et al*., 2014). The present study validates the above finding, *Oscillatoria* sp. was observed both in post pokkali season and post prawn season. They were absent during active prawn growth. Two *Fragillaria spp.* were seen in post pokkali season, prawns

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might have checked their growth. *Cymbella affinis* was reported from both plots during post pokkali season indicating that it may be a preferred diatom feed for prawns. In the present study nitrogen fixing blue-green algae *Oscillatoria sp., Anabaena spp.* were reported both from post pokkali season and post prawn season. This reveals that the algae present in the pokkali fields also have a significant role in the development of rice crops

CONCLUSION

The present work is to study the seasonal algal variation and water quality parameters of pokkali and prawn fields of Pizhala island. Diatoms were dominating in all seasons. Blue-green algae such as *Oscillatoria sp.* and *Anabaena sp.* were reported in post pokkali seasons. These nitrogen-fixing algae fix the atmospheric nitrogen which benefits the rice crops in the next pokkali seasons. It is evident that algae have great ecological significance in pokkali fields for the development of prawns and rice crops. The present study highlights the algal significance of pokkali fields and the need for their conservation. There should be birth for all vanished pokkali fields. Through the destruction of pokkali fields,much aquatic flora and fauna are also destroyed as well many traditional practices remain only in history textbooks.

ACKNOWLEDGMENT

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Table 1 List of Algal Flora Encountered For Study

Sl No.	Species	Post Pokkali season		Middle Prawn season		Post Prawn season	
		Plot 1	Plot 2	Plot 1	Plot 2	Plot 1	Plot 2
	Class Chlorophyceae						
1	*Oedogonium sp.*	+	+	-	-	-	-
2	*Spirogyra sp.*	+	+	-	-	-	-
	Class Cyanophyceae						
3	*Oscillatoria sp.a*	+	+	-	-	+	+
4	*Oscillatoria sp.b*	-	-	-	-	+	+
5	*Anabaena sp. a*	+	+	+	+	-	-
6	*Anabaena sp. b*	-	-	+	-	-	-
	Bacillariophyceae						
7	*Thalassiosira weisflogii*	-	-	-	-	+	-
8	*Skeletonema costatum*	-	-	-	-	+	+
9	*Fragilaria sp. a*	-	+	+	-		
10	*Fragilaria sp.b*	-	-	-	-	+	+
11	*Fragilaria sp.c*	-	-	-	-	+	-
12	*Tabellaria sp.*	+	-	-	-	-	-
13	*Eunotia minor*	-	-	+	+	-	-
14	*Cymbella affinis*	-	-	-	-	+	+
15	*Cymbella helvetica*	-	-	-	+	-	-
16	*Encyonema lacustre*	-	-	-	-	+	-
17	*Encyonema prostratum*	-	-	-	-	+	+
18	*Gomphonema sp.*	+	+	-	-	-	-
19	*Achnanthes inflata*	-	+	-	-	-	-

20	*Achnanthidium lanceolatum*	-	-	-	-	-	+
21	*Pinnularia divergens*	-	-	-	-	+	-
22	*Diploneis elliptica*	+	+	-	-	+	+
23	*Diploneis ovalis*	+	+	-	-	-	+
24	*Diploneis smithii*	-	-	-	+	-	-
25	*Navicula cincta*	+	+	+	-	-	-
26	*Navicula halophila*	-	-	+	+	-	-
27	*Navicula lanceolata*	+	+	-	-	+	+
28	*Navicula phyllepta*	+	+	+	-	-	-
29	*Navicula radiosa*	+	+	-	-	+	-
30	*Navicula tripunctata*	+	+	-	-	+	+
31	*Navicula viridula*	+	+	-	-	+	+
32	*Navicula sp. a*	+	-	-	-	-	+
33	*Gyrosigma acuminatum*	-	-	-	-	-	+
34	*Gyrosigma sp. a*	-	-	-	+	-	-
35	*Gyrosigma sp.b*	-	-	+	+	-	-
36	*Amphora coffeaeformis*	-	-	+	+	+	+
37	*Amphora ovalis*	-	-	+	-	+	+
38	*Amphora sp. a*	-	-	+	-	-	-
39	*Amphora sp. b*	-	-	+	+	-	-
40	*Bacillaria paradoxa*	+	+	+	+	+	+
41	*Tryblionella acuminata*	-	-	-	-	+	+
42	*Tryblionella angustata*	-	-	+	+	-	-
43	*Tryblionella gracilus*	-	-	-	-	+	-
44	*Nitzschia closterium*	-	-	-	-	+	+
45	*Nitzschia commutata*	-	-	-	-	-	+
46	*Nitzschia dissipata var. major*	-	-	+	+	-	-
47	*Nitzschia obtusa*	+	+	-	+	+	-
48	*Nitzschia ovalis*	-	+				
49	*Nitzschiapalea var. major*	+	+	+	+	+	+
50	*Nitzschia panduriformis*	-	-	-	-	+	+
51	*Nitzschia recta*	-	-	-	-	-	+
52	*Nitzschia sigmoidea*	+	+	-	-	+	+
53	*Nitzchia sigma*	-	-	-	-	+	+
54	*Nitzschia sp. a*	-	-	-	-	-	+
55	*Nitzschia sp. b*	-	-	-	-	+	-
56	*Coscinodiscus radiatus*	-	-	-	-	+	+
57	*Paralia sulcata*	-	-	-	-	-	+
58	*Surirella sp. a*	-	-	-	-	+	+
59	*Surirellasp.b*	-	-	-	-	+	+
	Class Euglenineae						
60	*Astasia fustis*	+	+	-	-	+	+
61	*Euglena sp.*	+	+	-	-	+	+
62	*Phacus sp.*	+	+	-	-	+	+

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Table 2. Physico- chemical variations of water in different pokkali seasons

Parameters	Post Pokkali season		Middle Prawn season		Post Prawn season	
	Plot 1	Plot 2	Plot 1	Plot 2	Plot 1	Plot 2
Temperature (degree Celsius)	27.5	27	27	28	32	31
pH	7.1	7	7.6	7.9	8.2	8
Alkalinity (CaCO3 mg/L)	55	50	100	90	125	120
Salinity (mg/L of NaCl)	1285.9	1004.9	19096.6	18337.6	21140.8	21841.6
Nitrite (mg/L)	2.8	3.2	3.4	3.7	2.2	2.9
Phosphate (mg/L)	4.4	3.4	4.6	3.8	3.3	3.4
Sulphate (mg/L)	150.19	262.54	1409.14	1368.85	1498.45	1515.63
Silicate (mg/L)	2.88	3.58	8.91	8.26	7.26	8.86
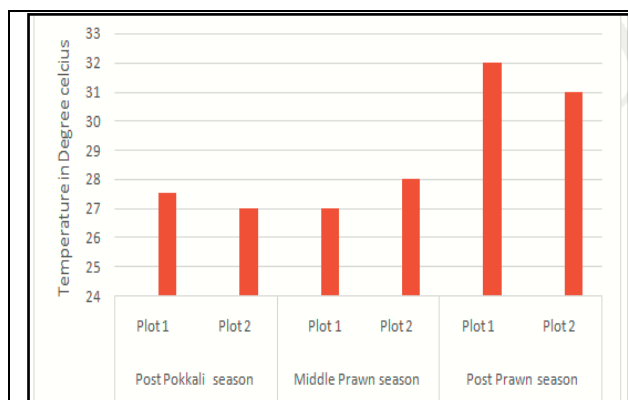
Figure 1: Graphical representation of temperature among three seasons
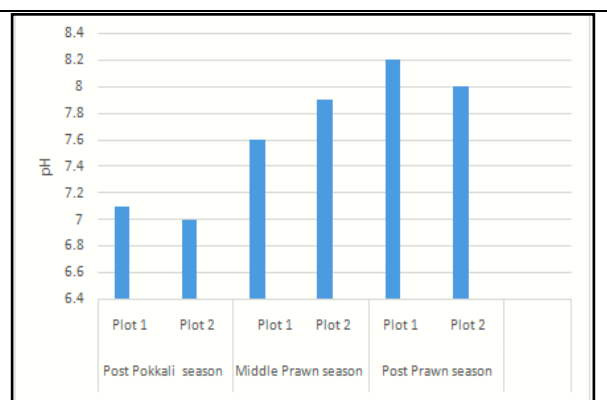
Figure 2: Graphical representation of PH among three seasons
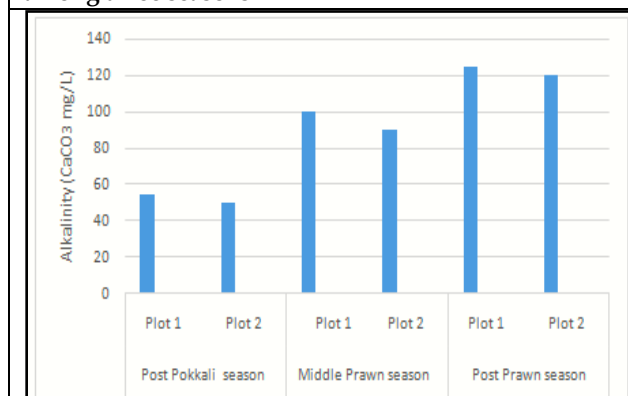
Figure 3: Graphical representations of variation of Alkalinity among three seasons
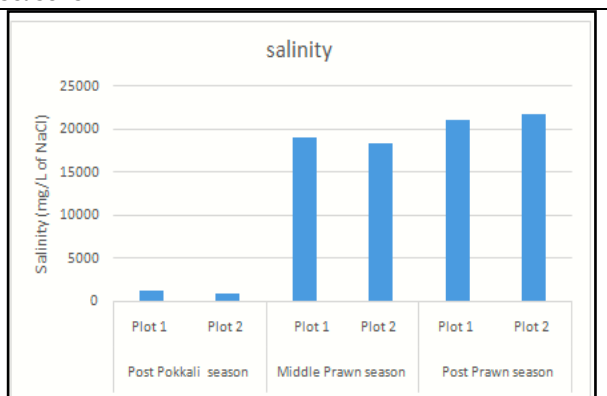
Figure 4: Graphical representation of salinity variations among three seasons

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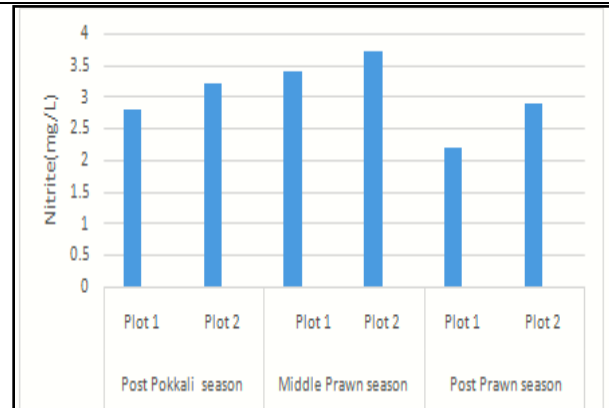
Figure 5: Graphical representation of Nitrite variations among three seasons
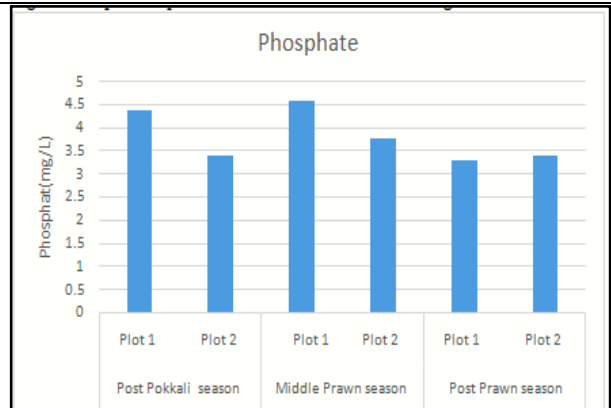
Figure 6: Graphical representation of Phosphate variation among three seasons
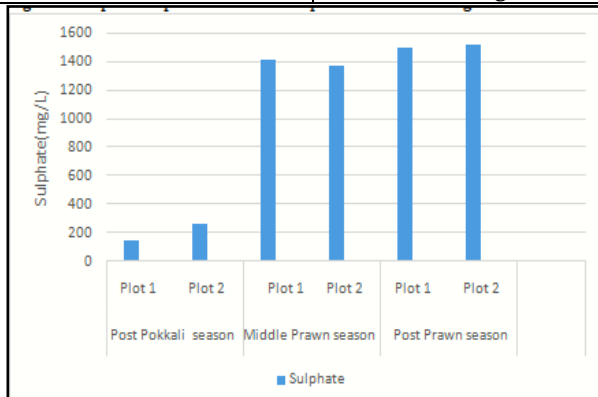
Figure 7 : Graphical representation of sulphate variations among three seasons

RESEARCH ARTICLE

Magnetic Nano Particle in Phenolic Waste Water Treatment

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ABSTRACT

The synthesized nano particles are used to treat the non-biodegradable and toxic organic matters. The sample treatment was carried out using Jar test apparatus, and the sample was treated in different time intravels. The magnetic nano particle was first used to treat the organic pollutant like chemicals O-Cresol, 2,6-Chlorophenol, and 2-Dichlorophenol and the percentage of removal is observed using UV visible spectrum by the optical density method. For this study, we use Al_2O_3 nanoparticles as a standard.

Keywords: water treatment, phenolic effluent, UV, Magnetic Nanoparticles, adsorption.

INTRODUCTION

Water purification using nanofiltration technology or through adsorption and catalytic degradation processes was made possible by the advances achieved and mysteries revealed in the quantum world [5]. Worldwide, the need for clean water is increasing because of population increase, drought and the contamination of conventional water sources. WHO (2004) reported that 1 billion people are at risk because they do not have access to potable water and another 2.6 billion people lack access to clean water [10,11]. The synthesis of magnetite nanoparticles has been intensively developed not only for its fundamental scientific interest but also for many technological applications: such as magnetic resonance imaging, ferro-fluids for audio speakers, magnetic targeted drug delivery, and magnetic recording media [1,8,9]. In particular, the use of magnetite nanoparticles as adsorbents in water treatment provides a convenient approach for separating and removing the contaminants by applying external magnetic fields [7].

Experimental procedure

The test water treatment is made in 250 ml beaker, 100 ml of mixed phenolic effluent is taken in the beaker, 2 mg of magnetic nano particles (γ-Fe_2O_3 doped with Ti, γ-Fe_2O_3 doped with Mg, $Ca(OH)_2$, Al_2O_3)   [2,3]added to it in room temperature. Beaker shacked well for equal distribution of magnetic nano particle. The external magnetic field is applied and the nano particle are immobilized 2 ml of the treated sample is taken into test tube and stock solution is added to it the color change takes place. The optical density is measured using UV spectrum. The procedure is repeated in various time intervals, Observations are listed in the table. Mixed wastes are collected and also used for test water treatment [3].

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RESULTS AND DISCUSSION

The water after treatment is taken to UV- spectrometer optical density analysis of their maximum absorption range. The effluent concentration was calculated from the standard calibration curve.

Effect of Time: Compare to Al_2O_3, Fe_2O_3 doped with Ti gives good result in removal of mixed effluent it gives 64% removal of organic mixed effluent in pH-8. Because it's oxidation state is +6 and magnetic property also high compare to other particles. After this Mg doped magnetic nano particle gives 62% effective removal, because it has less magnetization compared to Ti doped material. Al_2O_3 and $Ca(OH)_2$ gives 54% and 57% ,respectively. Both make complex material with effluent and settle it down. When the time increased with constant pH, the removing percentage is decreased, because the effluent particles make a layer and decreased the magnetization, so the effect of absorption also decreased. When increased the time, the removing percentage is decreased to 67-49%. Because absorbed effluent particles make a layer over the magnetic nano particles, this layer act as non-conducting material and block the magnetization this decrease the absorption. In Al_2O_3 and $Ca(OH)_2$ the formation of complex product decrease the absorption rate.

CONCLUSION

The prepared nano particle was used to treat the synthesized organic waste water and the effect of time and pH were observed through a jar test. In this study, we used Al_2O_3 as a standard. When Fe_2O_3 doped with Ti is used for water treatment it gives 20% high removal for organic pollutant compared with the standard Al_2O_3. By increasing the retention time of the sludge particles, that one is surrounded by the magnetic nano particles, the field strength is gradually reduced. It leads to decreasing the removal of sludge particles; hence the retention time should be maintained. By increasing the pH, the concentration of OH ions increases, this increased negative charge neutralized the magnetic field of the nano particle it may decreased the efficiency of sludge removal. Hence optimum pH should be maintained to increase the sludge removal during magnetic nano particle process.

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Table 1: Shows Initial Time UV Analysis Data for Treatment Chart

Nano particles	OD	OD*dil	Final Concentration	Initial Concentration	%removal
Fe_2O_3 Ti	0.542	1.626	10.6973	30	64.34
Fe_2O_3 Mg	0.564	1.692	11.1315	30	62.89
Al_2O_3	0.689	2.067	13.5986	30	54.67
$Ca(OH)_2$	0.652	1.956	12.8684	30	57.10
EFFLUENT	0.449	4.49	29.5394		

Table 2: Shows After 15 Min UV Analysis Data for Treatment Chart

Nano particles	OD	OD*dil	Final Concentration	Initial Concentration	%removal
Fe_2O_3 Ti	0.952	1.904	12.5263	30	58.24
Fe_2O_3 Mg	0.973	1.946	12.8026	30	57.32
Al_2O_3	1.258	2.516	16.5526	30	44.82
$Ca(OH)_2$	1.052	2.104	13.8421	30	53.85
EFFLUENT	0.449	4.49	29.5394		

Table 3: Shows After 60 Min UV Analysis Data for Treatment Chart

Nano particles	OD	OD*dil	Final Concentration	Initial Concentration	%removal
Fe_2O_3 Ti	0.771	2.313	15.2171	30	49.27
Fe_2O_3 Mg	0.798	2.394	15.75	30	47.5
Al_2O_3	0.902	2.706	17.8026	30	40.65
$Ca(OH)_2$	0.817	2.451	16.125	30	46.25
EFFLUENT	0.449	4.49	29.5394		

Table 4: Shows UV Analysis Data for Treatment Chart

Nano particle Time	Fe_2O_3 Ti	Fe_2O_3 Mg	Al_2O_3	$Ca(OH)_2$
Initial Minutes	64.34	62.89	54.67	57.10
15 Minutes	62.45	60.57	50.87	55.26
30 Minutes	58.24	57.32	44.82	53.85
60 Minutes	49.27	47.5	40.65	46.25
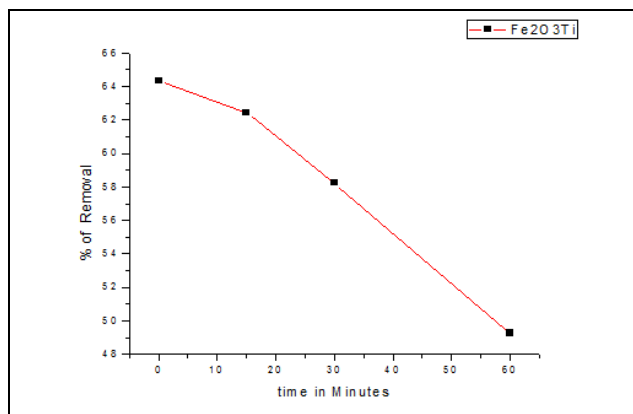
Fig 1: Effect of time in phenolic effluent for Fe_2O_3 doped with Ti	**Fig 2: Effect of time in phenolic effluent for Fe_2O_3 doped with Mg**
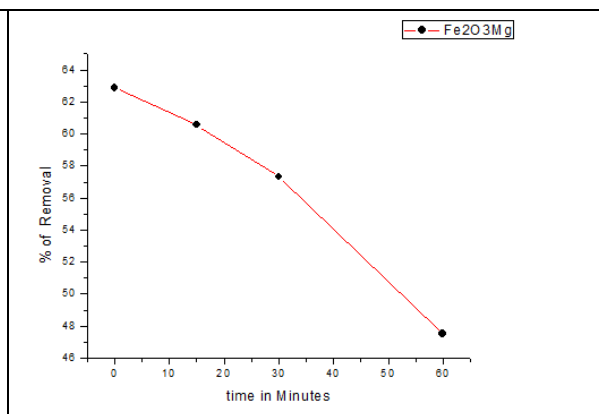
Shanmuga Selvan *et al.,*
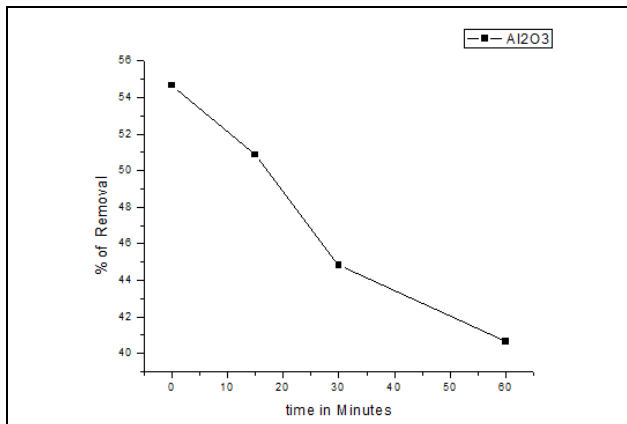
Fig 3: Effect of time in phenolic effluent for Al₂O₃
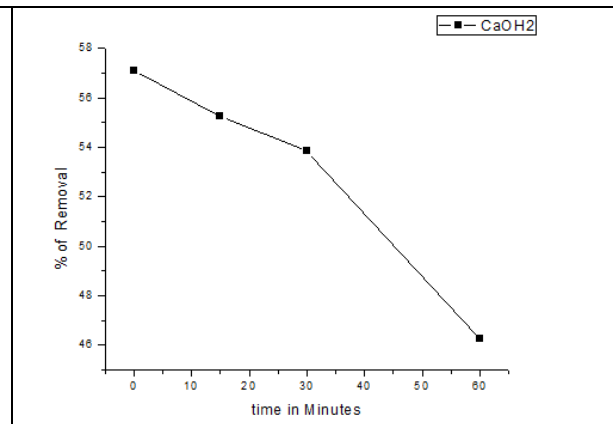
Fig 4: Effect of time in phenolic effluent for Ca(OH)₂
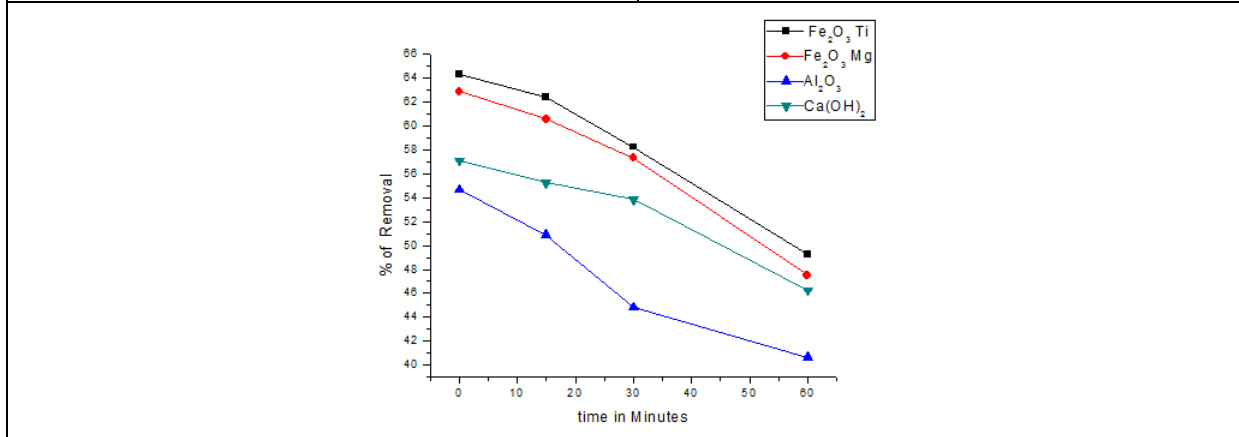
Fig 5: Effect of time in phenolic effluent

RESEARCH ARTICLE

IoT-based Human Activity Classification using Convolutional Neural Network and Metaheuristic Technique

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ABSTRACT

The Internet of Things represents the fastest flourishing technology paradigm containing plenty of devices that produce, process and communicate large amounts of data. The Internet of Things (IoT) is a technology paradigm that combines sensing, actuation, communication, compute, storage, and networking to deliver essential information to end-user systems and applications. IoT has a broad study scope across all areas such as implementation, architecture, processing of data, and standardizations because it is a newfound sector in terms of technology and applications. Diverse human activities are considered, and the activities are classified with the assistance of a convolutional neural network (CNN). The classification accuracy is enhanced by the process of feature selection that is done using hybridized linear discriminant analysis (LDA) based genetic algorithm (G.A.). The results acquired from the classification approach are investigated with performance metrics, and the numerical outcomes are compared with existing state-of-art techniques. The proposed LDA-GA-CNN-based classification approach outperforms the existing classification techniques.

Keywords: Classification, sensor, neural network, human activity, fitness function, accuracy and activity prediction

INTRODUCTION

The Internet of Things (IoT) has acquired a lot of attention presently. It is a fascinating technology that offers many answers to challenges that arise in many sectors. The Internet serves as a network's communication backbone for exchanging information from one system to another throughout the world. The Internet of Things (IoT) consists of a billion linked gadgets that can sense, collect, and transport data from one device to another without the need for human intervention. Health care monitoring, intelligent buildings, logistics, connected autos, innovative city

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development, smart grid, smart home, smart retail, smart agriculture, and other applications of IoT are only a few instances. The IoT is fast evolving from a theoretical notion to practical reality, thanks to WSN, sensors, and gateways with cloud platforms [1]. The sharpness of technology is enhanced by developing both vital components, which includes numerous advancements in sensing elements, data fusion, aggregation, refinement, and collecting. Aside from that, advances in data management on cloud platforms have aided in the growth of IoT. The perception layer gathers data, the network layer for establishing connectivity, the middleware layer with its sub-layers for data handling, models, software/tools,and platforms are all part of the IoT system architecture [2, 3].

Traditional processes may not give better data for monitoring the environment than a thing. The IoT is being used to monitor and manage natural disasters and human activities. This mechanism collects data using sensors such as an ultrasonic sensor, a rain sensor, a temperature sensor, and a humidity sensor. It then transmits the information from one end to the other. The irrigation, monitoring, or disaster management departments may use an IoT system to centrally monitor, evaluate, and forecast essential conditions [4]. The forecasting method can warn the administration and the public about the possibility of a decline. In the future, this will aid in the implementation of required actions to reduce the number of lives lost and property damage [5, 6]. Sensor data from various motions and locations is collected and used by the system. This system collects sensory data and displays it on a central dashboard in real-time—a predictive method for predicting the likelihood of a fall after it has occurred [7]. By providing precise fall risk estimates, this approach has the potential to save the elderly and citizens' lives [8, 9].

The IoT is a network of battery-powered devices or things that may be linked to the web in various ways. The most common means of communication is wireless, but cable transmission is also available and suited for some purposes [10]. Data storage, transport, and processing are necessary components of an IoT system. Various technologies are used in IoT development at different stages, including edge devices that can sense and send data. Data transfer is enabled through connectivity technologies, which use a variety of protocols to understand the recovered data. The data management platform offers data storage capacity, whereas the application platform enables services that allow the main program to integrate with device data libraries. Edge, Network, and Core are the three fundamental components of IoT. The edge is concerned with things directly, whereas the core is concerned with massive data, and the network is concerned with diverse communication techniques. Edge devices may convert to fog or use networking technologies to deliver data to the cloud. The data collected can be saved and processed at the edge, in the fog, or the cloud [12]. The research article is systematized as follows: the literature review is given in Section 2, the activity classification methodology is provided in Section 3, the outcome of the proposed technique is illustrated in Section 4, and the research is concluded in Section 5.

Related Works

Esfahani *et al.* demonstrated that adding many sensors across properly nominated locations on the individual can considerably improve the precision of identifying the individuals' actions [13]. The task entailed gathering data from people and then putting it in a pool to see where it belonged. Detecting complicated behaviors would be a massive challenge since a new model would be required; most untrained data can be categorized as predicted. SVM and its relevant classifiers and the piece-wise linear SVM (PL-SVM) [14] are used extensively in computer vision for object surveillance. Because it may quickly give helpful discernment into any moving object, the approach is commonly used to analyze and recreate from two-dimensional perspectives. This study highlights the necessity to use this attribute concisely. Still, the main disadvantage is that it will not readily give a solution if we need to define activities more accurately.

Smartphone sensors can monitor human activity since they are ubiquitous and allow simple access to essential data. Intelligent sensors, according to Ahmed et al., are more qualified than the traditional methodology of streaming video [15].However, sensors that reside in smartphones becomes convenient. Chen et al.'s research show that raw data needs to be processed with more technology to provide a pleasant result [16]. That is why, in the study, we have placed a greater emphasis on the utilization of efficient hardware-based classifiers, divining the data in training and

test data, and extracting features to avoid a gap among performance and sensor data. Mohammed Mehedi Hassana *et al.* used wearable sensors in a smartphone to capture human physical activities in signal across three axis of a gyroscope and accelerometer. After collecting features from raw data to improve their strength, kernel principal component analysis and LDA is used to analyze them. Finally, a deep belief network trains features (DBN). The results of the DBN were then compared to those of classic expression recognition methods such as the multiclass SVM and artificial neural network (ANN). On the test set, they achieved accuracy of up to 95.85% [17].

RubénSan-Segundoa *et al.* used a three-axis gyroscope and accelerometer to collect data from smart phones and smart watches. Two approaches were used to extract features from the raw data: HMM and CNN. The accuracy of the smart watch increased from 78.6 to 88.1, while the accuracy of the smart phone increased. This is clear evidence that feature normalization is helpful for smar tphone data but not for data from smart watches [18]. Data were collected from three separate sensors by Ran Zhu et al. The sensory samples of 100 people made up the dataset. They used a convolutional neural network to handle the dataset and got a 95.62 percent accuracy while employing unique ensembles of CNN gave a 96.29 percent accuracy [19]. Tsige Tadesse Alemayoh *et al.* utilized a smart phone to translate accelerometer and gyroscope motion sensors into a 14 60 virtual picture. They then created an iOS app for capturing and broadcasting motion data and identifying real-time activity. These processed data were sent into the specified convolutional neural network for classification. The time-series data format can produce superior outcomes in terms of accuracy for forecasting activities. They achieved an overall accuracy of 99.5 percent [20].

SVM, K-NN, and logistic regression were utilized in retrieving features from raw data by Li Fang et al. SVM had an average accuracy of 88.9%, KNN had an average accuracy of 95.3 percent, and logistic regression had an average accuracy of 83.9 percent. The focus of the piece was primarily on up and down buses. SVM was 89.5 percent accurate for up bus, KNN was 96.8% valid, and L.R. was 89.7% accurate [21]. SVM was 84.2 percent accurate for the down busrate, KNN was 94.2 percent accurate, and L.R. was 81.9 percent accurate [21].

Proposed Methodology: Classification using LDA-GA-CNN
Feature Selection-LDA-GA
LDA works to classify the samples effectively. The LDA process is used as a fitness function in a genetic algorithm. Initially, compute the mean value (μx) for each feature in feature space and compute the covariance matrix (cx) of each population. The covariance is estimated as.

$$c_x = \sum_{i=1}^{n} (X_i - \bar{x})(Y_i - \bar{y})/(n-1)$$

where $\bar{x}, \bar{y}$ is the mean of X and Y. The group covariance matrix is calculated as

$$C = \frac{1}{n}\sum_{n=1}^{g} n_x c_x$$

wherenx is the count of examples in the population, x, g is the count of populations, vx is the initial probability vector. The prior probability vector is computed

$$V_x = \frac{n_x}{g}$$

After computing the covariance matrix, the minimum discriminant value is derived as

$$d_x = \mu_x C^{-1} a_i^T - \frac{1}{2}\mu_x C^{-1}\mu_x^T + ln v_x$$

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where dx is denoted as the minimum discriminant value, ai is the feature array in sample I. The sample can be allocated to population x, with minimal discriminant dx. Based on these computed values, the genome fitness value is estimated as follows,

$$F1(\beta) = M^{-1}(\mu_1 - \mu_2)$$
$$M = \frac{1}{(n_1 + n_2)}(n_1 M_1 + n_2 M_2)$$

Calculation of linear co-efficient maximizes the feature score that is estimated according to the following equation. Linear co-efficient model is indicated as β, matrix of covariance is indicated as M and average of the feature vector is indicated as μ. The Mahalanobis (Δ) is utilized to estimate discriminants among a group of vectors. After computing fitness value, the number of genes has been selected as.

$$Number of selected gene F2(I) = \frac{1 - m_r}{p}$$

where m_τ is denoted as the count of bits having the esteem "1", p indicates the length of the chromosome. From the processed affection, the capability is evaluated as.

$$F(I) = \alpha f_i(I) + (1 - \alpha)f_2(I) subject to\ 0 < \alpha < 1$$

Let α be more prominent than 0.5 for high characterization precision and lesser than 0.5 for tiny measured quality subsets. After computing the fitness value of each individual in the feature space, the roulette wheel selection approach is applied to select the solution for performing the recombination process. The computed fitness value is assigned to the chromosomes based on the fitness value; the probability value of selection is estimated to select the chromosomes from the features set. The probability value is computed as.

$$P_i = \frac{f_i}{\sum_{j=1}^{N} f_j}$$

where N is the whole count of individuals in feature space, fi is the fitness value computed according to F(I). As per the likelihood esteem, the enhanced features are chosen, which is more survived in search space compared to weaker ones. The repeated individual feature value has better survival in feature space than other features used to generate the next new offspring.

The effective probability selection process reduces the weakest feature and also improves the survival rate of the highest probability fitness feature. A uniform crossover process is applied from the selected element to generate the new offspring. During this process, each bit is chosen independently from the parent chromosome. Then the selected points are interchanged for developing the new offspring, but it does not require bias and segmentation. Finally, the inverse mutation process is applied to the selected crossover feature. The generated components are replaced by the existing part in the population list. This process is repeated until it reaches the termination condition. In this work, the features selection process is performed to compare the 10N epoch. The selected features are denoted as one, and eliminated features are represented as zero. The collection of subsets is created by every genotype based on its phenotypes. These subsets are utilized as preparing sets.

Classification

The last step of the work is activity classification which is done by applying N.N. The network is simply a radial basis convolutional neural network with a twist. The network operates using a nonparametric regression procedure, with the training samples designated by the radial neurons' mean. The random function is computed in the input and output by the neural network. Based on the discussion, the general arrangement of the system appears in Figure

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1. The above image depicts the structure of N.N. that is made up of three layers: input, output, and convolutional. The needed knowledge from the selection phase is supplied to the input phase, and output is analyzed and estimated by the output phase. Each phase performance is specifically determined as.

$$E[a|b] = \frac{\int_{-\infty}^{\infty} a * f(a,b)dy}{\int_{-\infty}^{\infty} f(a,b)dy}$$

Where b indicates the variable of estimation for the output phase, aindicates the input value, E[a|b] indicate the predictive value of output specified the input vector b, f(a, b) indicates the Joint Probability Density Function (PDF) of a and b. To obtain the final output, the calculated value is sent to the output units that are specified, and the output estimation variable is defined explicitly as,

$$y_i = \sum_{i=1}^{n} h_i w_i j \sum_{i=1}^{n} h_i$$

where wij indicate the target yield relating to including preparing vector xi, $h_i = e^{(\frac{1-D_i^2}{2*\sigma^2})}$is the output of a hidden layer neuron, $D_i^2 = (a - UI)T(a - UI)$ is the squared distance amongst the input vector a and the training vector u; a is the input vector, U.I. is training vector, smoothing factor σ = a constant controlling the size of the receptive region. The incoming activity features are successfully analyzed and classified with a minimum execution time and minimum error of a different smoothing factor.

RESULT AND DISCUSSION

This section discusses the results acquired from diverse existing techniques and LDA-GA-CNN proposed approach. Analysis of results and discussion about the results are illustrated in detail.

Dataset Description
The legitimacy of the approach described here is tested using a simulated dataset. In the feature evaluation phase, an ideal feature vector is constructed based on the feature identification, which is then employed in the classification process during neural network-based human action recognition. These axial data were used to compute features. A strong heterogeneous feature vector is derived from the time-series data to reduce the likelihood of incomplete details. 138 characteristics are taken from the data of the six-axial sensor to detect the activity, even though there are 23 original features.

Performance Metrics
Accuracy: Accuracy is defined as the closeness of the determined truths from the categorized instances, prediction of the actual value, and recognition of both T.P. and T.N. values amongst the count of assessed classes.

$$Accuracy = \frac{TruePositive + TrueNegative}{TruePositive + TrueNegative + FalsePositive + FalseNegative}$$

Sensitivity: The rate of hit rate, true positive, and recall are all indicators of sensitivity, defined as the portion of positive values that are accurately detected. It's computed as follows:

$$Sensitivity = \frac{TruePositive}{TruePositive + FalseNegative}$$

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Specificity: The ratio of genuine negative values out of all the collected samples is known as specificity, and it has no precise condition. It is estimated as,

$$Specificity = \frac{TrueNegative}{TrueNegative + FalsePositive}$$

Precision : The positive quantitative value, also known as precision, denotes the reliability of the measurement and the importance of the recognized values. Random mistakes are defined as precision, which is calculated using statistical factors. It is estimated as,

$$Precision = \frac{TruePositive}{TruePositive + FalsePositive}$$

Matthews correlation coefficient: MCC is used in machine learning systems as a criterion for classification quality. The correlation coefficient between the predicted and observed categorization values are examined. It's computed as follows:

$$MCCValue = \frac{TP \times TN - FP \times FN}{\sqrt{(TP + FP)(TP + FN)(TN + FP)(TN + FN)}}$$

Error rate: The data transformation, caused by numerous causes like noise, distortion, and interference, is the source of the mistake. The error rate is defined as the fraction of sequences miscategorized by the decision-making model as a ratio of a performance rate. It's computed as follows:

$$Errorrate = \frac{FP + FN}{TP + TN + FP + FN}$$

From the observation of performance, it is identified that the proposed LDA-GA-CNN is highly effective in terms of performance metrics. It achieved high accuracy with a minimal error rate.

CONCLUSION

The Internet of Things (IoT) is a rapidly growing technological paradigm that includes many devices that create, analyze, and transfer massive amounts of data. The Internet of Things (IoT) is a technology paradigm that combines sensing, actuation, communication, computing, networking, and storage to deliver real-world data to end-user applications and systems. IoT has a broad study scope in all areas such as architecture, implementation, data processing, and standardization because it is a new sector in technology and applications. Diverse human behaviors are taken into account, and the actions are categorized using a convolutional neural network (CNN). The feature selection procedure, which uses a hybridized linear discriminant analysis (LDA) based genetic algorithm (G.A.), improves classification accuracy. The proposed approach attains 99% accuracy with a minimal error that shows the effectiveness of the proposed technique. The system can be extended with other deep learning techniques in the future.

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Table 1. Comparison of Accuracy

Epoch	SVM	GA	CNN	LDA-GA-CNN
50	79	84	88	90
100	81	85	89	91
150	81.5	86	90	92
200	82	88	91	96
250	83	89	91	99

Table 2. Comparison of Sensitivity

Epoch	SVM	GA	CNN	LDA-GA-CNN
50	77	80	84	89
100	78	81	85	90
150	78.5	82	86	91
200	79	83	86	93
250	81	83.5	88	93.5

Table 3. Comparison of Specificity

Epoch	SVM	GA	CNN	LDA-GA-CNN
50	78	79	83.5	90
100	79	80.5	84	91
150	79.5	81	85	92
200	80	82	86	93
250	81	83	87	94

Table 4. Comparison of Precision

Epoch	SVM	GA	CNN	LDA-GA-CNN
50	78	81	82	85
100	79	83	83	86
150	81	83.5	85	89
200	81	84	86	92
250	81.5	86	87	93

Table 5. Comparison of MCC

Epoch	SVM	GA	CNN	LDA-GA-CNN
50	78	79	81	90
100	80	80	84	91
150	81	81	86	93
200	83	83	87	95
250	84	85	88	96

Table 5. Comparison of Error Rate

Epoch	SVM	GA	CNN	LDA-GA-CNN
50	7.54	6.7	6.2	4.1
100	7.76	6.8	6.1	4.6
150	7.9	6.9	5.9	4.7
200	8.1	7.2	6.2	5.1
250	8.2	7.4	6.5	5.2

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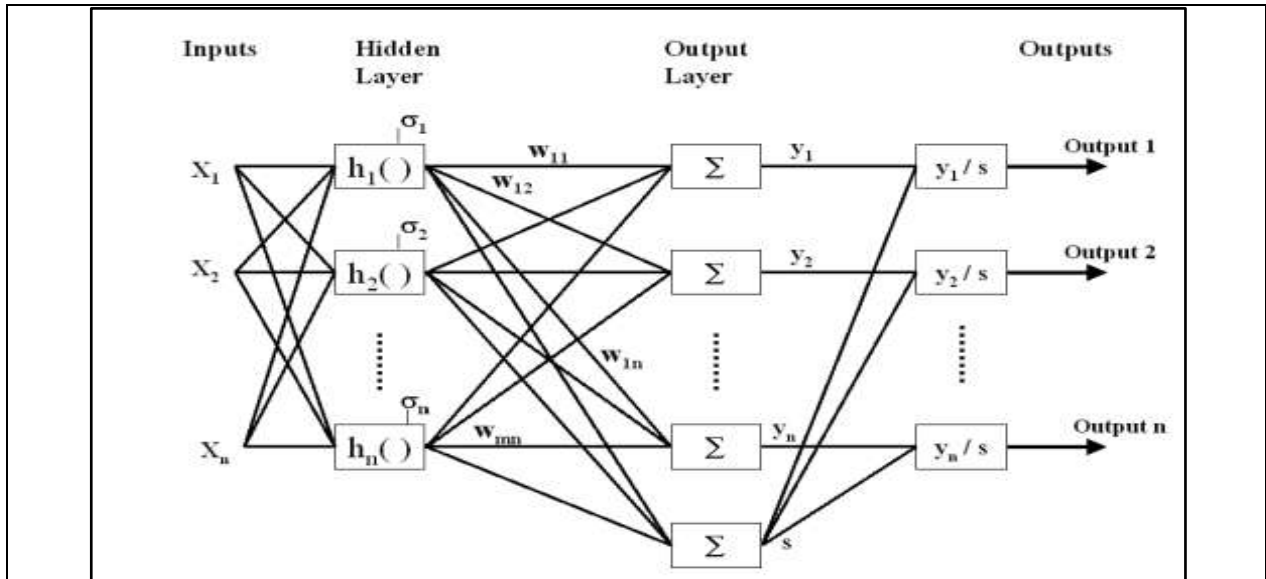
Figure 1. The overall structure of N.N
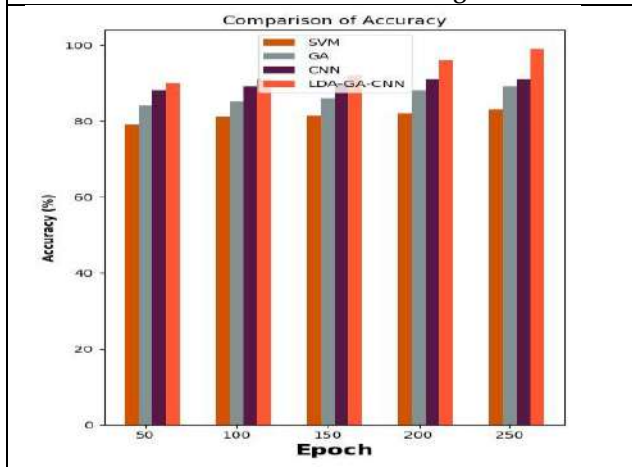
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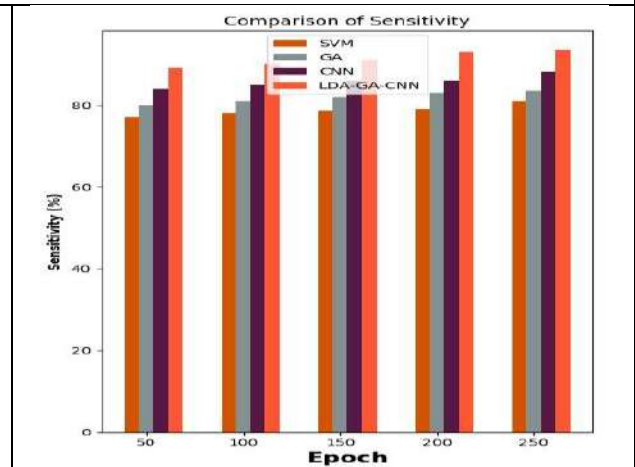
Figure 3. Comparison of Sensitivity
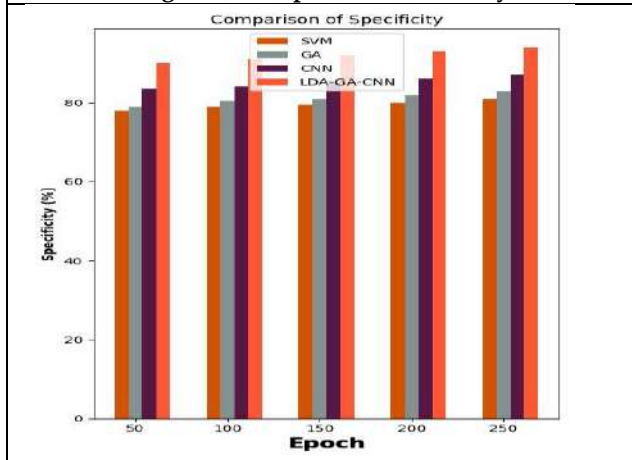
Figure 4. Comparison of Specificity
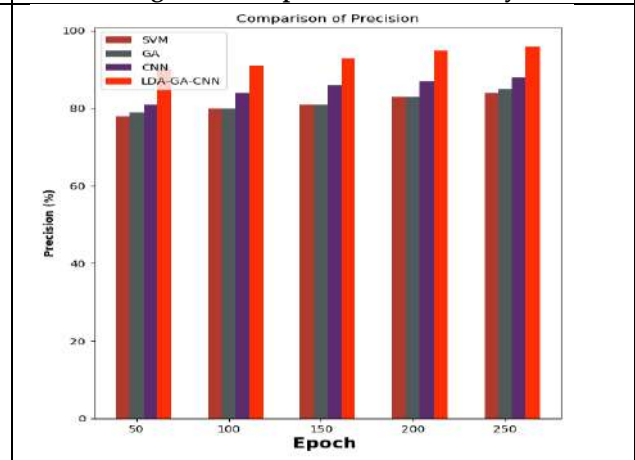
Figure 5. Comparison of Precision

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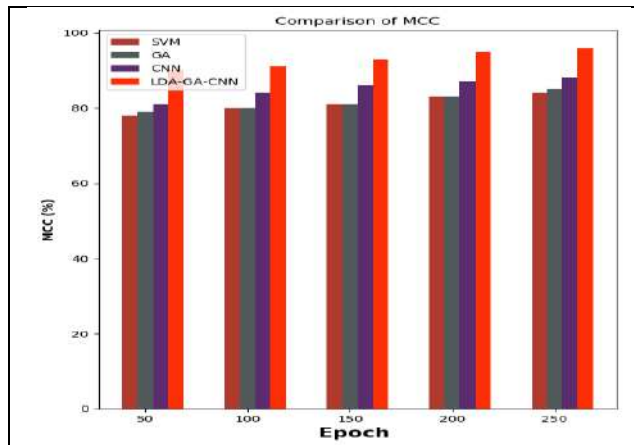
| Figure 6. Comparison of Precision | Figure 7. Comparison of Error Rate |
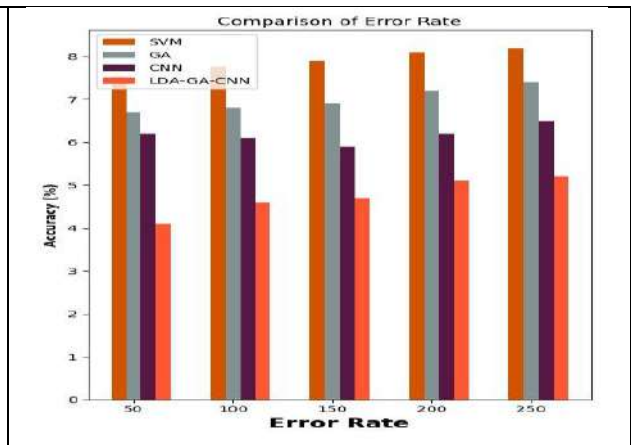
RESEARCH ARTICLE

Synthesis, Characterization and Antimicrobial Screening of Some Novel 5-nitro [1] Benzofuropyridinone and their Derivatives

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ABSTRACT

Encouraged by the pharmaceutical properties of benzofuran compounds and in continuation of our synthetic work on pharmacologically potent benzofuran derivatives, we now report the synthesis of novel derivatives of benzofuran linked with pyridinone compounds. The desired compounds were prepared by using 2-nitrosalisaldehyde oxime (1). Compound 1 on treatment with acetic anhydride to produce corresponding nitrile (2), which on cyclisation with chloroacetone results in the formation of 1-(3-amino-5-nitro-1-benzofuran-2-yl)ethan-1-one (3). The compounds 4(a, b) produced by acetylation and benzoylation reaction respectively. On alkylation of compounds 4(a, b) we get new pyridinone compounds 5(a-b). All the new heterocyclic compounds synthesised were characterized by IR, ^{1}HNMR and GCMS spectral data. Further, these have been screened for their antimicrobial, antifungal activity.

Keywords: Benzofuran, pyridinone, acetylation, benzoylation, Antimicrobial, Antifungal activity.

INTRODUCTION

Since long time we are working on the synthesis of various benzofuran derivatives of biological interest, which have been reported a wide spectrum of biological and pharmacological activities. The benzofuro pyridinone ring 1is the essential feature of a large number of biologically active compounds namely penicillins, cephalosporins, carumonam, aztreonam, thienamycine and the nocardicins. Recently, besides the antibacterial activity, antifungal, antitubercular, antitumor, cholesterol absorption inhibition and enzyme inhibition activity have been reported in compounds containing 2-azetidinone ring systems.[1-4]. Due to their relevance in both clinical and economic fields, various

substituted 2-azetidinones represent a very attractive target for contemporary organic synthesis [5-10]. The various benzo [2, 1-*b*] furan derivatives synthesized in this laboratory have shown to possess wide spectrum of biological and pharmacological activities [11-16]. There are several evidences that the presence of nitro group enhance the biological activities of the Compounds many folds. Thus 5-nitrobenzofuran, 5-nitro-3-aminobenzo [2, 1-*b*] furan and 5-nitro-3-aminobenzo [2, 1-*b*] furan have been synthesized and evaluated for possible antimalarial and mutagenic activities and they also find application in veterinary medicine. Hence, with a view to further assess the pharmacological profile of this class of compounds[17-20], it was thought worthwhile to synthesize some new congeners of -lactam heterocycles by incorporating the benzo [2, 1-b] furan nucleus bearing nitro group and pyridinone moieties in a single molecular framework. The present work deals with the synthesis of the title compounds starting from 2-methyl-8-nitro[1]benzofuro[3, 2-*b*]pyridin-4(3*H*)-one, followed by their antifungal and antimicrobial screening [21].

MATERIALS AND METHODS

General Information
All the chemicals, reagents and solvents used obtained from commercial sources and analytical grade. All the reaction were monitored by thin layer chromatography (TLC) using silica gel as stationary phase, different solvent systems as mobile phase and uv chamber as detecting agent. Melting point was determined on an electronic apparatus and is uncorrected. IR spectra were recorded on Shimadzu model IR-435 spectrophotometer using KBr discs for solids and thin films for liquids. ^{1}H NMR spectra were recorded on Joel AC (400 MHz) in DMSO-d$_6$ and CDCl$_3$ using tetramethyalsilane (TMS) as an internal slandered. TOF ES+ Mass spectra (m/z) were recorded on micro mass Auto spec LCTKC455. Substituted benzofuran pyridinone were prepared by literature procedures, starting from the corresponding salicylaldehyde oxime and the other reactants. 2-methyl-8-nitrobenzofuropyridinone was prepared in five steps following the literature procedures.

Experimental Procedure
Preparation of 5-nitrosalicylaldehyde oxime (1)
A mixture of 5-nitrosalicylaldehyde (5g, 0.01mol) and hydroxylamine hydrochloride (2.8g, 0.017 mol) were dissolved in a mixture of freshly distilled ethanol and water. The solution continuously stirred and a solution of NaOH (5.2g in 5ml water) was added in portions, carefully avoiding vigorous reaction. After the completion of addition of NaOH, the reaction flask was cooled to room temperature. Then the reaction mixture was refluxed for 20 min. It was cooled and diluted with ice cold water. The separated unreacted solid 5-nitro-2-hydroxybenzaldehyde was removed by filtration. The filtrate was cooled and carefully acidified with dilute HCl. The precipitated oxime was filtered and washed with cold water and recrystallized from methanol. (Melting point=78^0 C,   yield: 82.38%)

Preparation Of 5-Nitro-2-Hydroxybenzonitrile (2)
The dried 5-nitrosalicyldehyde oxime **(1)** (20 g) obtained above was transferred to a round bottomed flask and added with anhydrous acetic anhydride (20 ml) was refluxed for 30 min.; excess of acetic anhydride was removed by distillation under reduced pressure. The dark dense liquid of benzo[1, 2-d]isoxazole was treated with freshly prepared sodium ethoxide [prepared by adding freshly cut dry sodium (20 g) into absolute ethanol (100 ml) at 0^0 C]. The mixture was stirred for 30 min. at room temperature and poured into ice-cold water. On acidification with HCl, it gave 5-nitro-2-hydroxybenzonitrile **(2)** as light brown solid, which was collected and recrystallized from ethanol (Melting point-94° C, yield: 62.5%)

Preparation Of 2-Actyl-3-Amino-5-Bromo-1-Benzofuran (3)
5-Nitro-2-hydroxybenzonitrile (**2**) (1.64 g, 0.01mol) was dissolved in dry acetone (50ml). To this solution, finely grounded anhydrous potassium carbonate (3g) and chloroacetone (1.10ml, 0.01mol) was refluxed for 12 hours. The potassium carbonate was filtered off and from the filtrate acetone was removed under reduced pressure. The resulting light brown solid was collected and recrystallized from ethanol. (Melting point: 114°C, yield: 68.2%)

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Preparation Of 3-Acyalaminobenzofuro [2, 1-B]-2-Corboxylate (4a, B)

The compound **3** (2.28 g 0.01 mol) was heated on hot water-bath(60-70⁰C) for 20 min with acetyl chloride (4 ml) in the presence of aqueous sodium hydroxide (2N, 2 ml). The product **4a** thus separated as solid was collected and recrystallized from ethanol. Similarly compound **4b** was obtained by treating compound **3** with benzoyl chloride**.** Yields, melting points and solvent used are given in table 01.

Preparation Of 2-Methyl-4-Oxo-Benzo [2, 1-B] Furo-[3, 2-D]Pyridinone (5a, B)

Acetyl compound **4a** (2.62 g 0.02 mol) was suspended in aqueous sodium hydroxide (1N 500ml) and warmed on a water-bath(60-70⁰C) for 30 min. The resulting solution when carefully acidified with dilute HCl gave compound **5a**, which was then purified by recrystallisation aqueous ethanol. The same procedure was followed to obtain compound **5b** from compound **4b.** Yields, melting points and solvent used are given in table 01.

RESULTS AND DISCUSSION

The synthesis of the titled compounds is depicted in the (Scheme). All the intermediate and targeted molecules were confirmed by TLC, IR, NMR and Mass spectral data. 2-acetyl-3-amino-5-nitro-1- benzofuran **(3)** was synthesized by treating 5-nitro-2-hydroxybenzonitrile **(2)** and chloroacetone in presence of anhydrous potassium carbonate as per the established procedure in this laboratory. 3-Acylaminobenzo[2,1-b]furan-2-corboxylate was prepared by stirring acetyl chloride **(4a)** and benoylchloride **(4b)** respectively in the presence of NaOH in cold condition. Compounds 2-methyl-4-oxo-benzo[2,1-b]furo-[3,2-d]pyridinone **5(a, b)** were obtained by refluxing in presence of sodium hydroxide. The IR spectra of the targeted molecule **5b** revealed the peak at 1645 cm-1 showing the Presence of C=O stretching vibration of cyclic amide. The C-H stretching frequency was at 2921 cm-1 indicated their presence in pyridinone ring. From ^{1}H NMR spectra of **5b** exhibited the signal at δ ppm 2.8 which was assigned to methylene (-CH$_2$-) protons and the multiplet signal δ ppm 7.15 to 7.69 were due to aromatic protons. The structure of the compound **5b** was further supported by its ESI MS Spectra indicating molecular ion peak was at 305 *m/z* and fragmented ion peak at 285 *m/z.*

Compound 2:
IR in cm^{-1}(KBr) : 3303 (OH-aromatic stretching), 2238(C≡N), 1535 (NO$_2$);
^{1}H NMR in δ ppm (DMSO): 8.19-8.21 (s, 1H, aromatic OH), 7.05-8.19 (m, 3H, Ar-H); Mass M$^+$ peak (m/z) : 165 (164) (M+1)$^+$.

Compound 3:
IR in cm^{-1}(KBr): 3452 (NH$_2$ stretching), 1658 (C=O), 1525 (NO$_2$); ^{1}H NMR in δ ppm (DMSO) : 6.17-6.14 (s, 2H, -NH$_2$), 2.33(s, 3H, CH$_3$), 8.46(s, 1H Ar-H), 7.78-8.09(m, 2H, Ar-H);Mass M$^+$ peak (m/z) : 219-220 (220).

Compound 4b:
IR in cm^{-1}(KBr) 3330(-NH-CO-C$_6$H$_5$), 1721(-C=O), 1665(-CO-NH-),1523(-NO$_2$); ^{1}H NMR in δ ppm (DMSO) : 2.55(s, 3H, -CH$_3$), 7.52(s, 1H, -NH-), 7.44-7.51(t, 2H, Ar-H), 7.41-8.11(m, 6H, Ar-H); Mass: M$^+$ peak (m/z) 324.99 (324 may be due to M+1)

Compound 5b:
IR in cm^{-1}(KBr) : 2921(CH$_2$ Ar-), 1645(-C=O), 1618 (C=N-), 1526(-NO$_2$);
^{1}H NMR in δ ppm (DMSO) 2.8 (s, 2H, -CH$_2$), 7.3(m, 3H, Ar-H), 7.6-8.1(d, 4H, Ar-H), 8.4(s, 1H, Ar-H); Mass: M$^+$ peak (m/z) 305-306(305 may be M-1 peak).

Biological Study
Antimicrobial Activity

The antimicrobial activity of newly synthesized compounds was determined by cup-plate method. The *in vitro* antimicrobial activity was carried out for old cultures of three bacteria and two fungi. The bacteria used were *Pseudomonas aureginosa*, *Escherichia coli* and *Staphylococcus aureus.* The fungi used were *Aspergillus Niger and*

Candida albicans. The compounds were tested at a concentration of 0.001 mg/ml in D*o derma* MF against both the organisms. The amoxicillin and fluconazole were used as standard for comparison of antibacterial and antifungal activities respectively. The zone of inhibition was compared with the standard drug after 24 h and incubation at 30^{0}C for antibacterial activity and 72 hours at 25 ^{o}C for antifungal activity. The compound **5b** showed promising activity against *Pseudomonas aureginosa, Escherichia coli* and *Staphylococcus aureus.* The compounds **4a** and **4b** exhibited moderate activity against *E. coli.* For antifungal activity, there was good zone of inhibition.

CONCLUSION

A series of novel 5-nitro-benzofuropiridinone have been synthesized in good yield and screened for their antibacterial and antifungal activities. In the series, the compounds bearing 5-nitro and carboxyl group exhibited potent activities compared remaining. However these *invitro* evaluations in different experimental models and detailed toxicological studies are necessary to further support these results. The detailed metabolic stability assay is still in progress.

ACKNOWLEDGEMENTS

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Table 01: Physical characterization data of synthesized compounds

Compound	R	Molecular Formulae	M.P ^{0}C	Yield (%)	Solvents
2	-	$C_7H_4N_2O_3$	94	62.5%	Ethanol
3	-	$C_{10}H_8N_2O_4$	114	68.2%	Ethanol
4a	CH_3	$C_{12}H_{10}N_2O_5$	113	59.5%	Ethanol
4b	C_6H_5	$C_{17}H_{12}N_2O_5$	162	56.8%	DMF
5a	CH_3	$C_{12}H_8N_2O_4$	120	61.3%	Ethanol
5b	C_6H_5	$C_{17}H_{10}N_2O_4$	112	63.8%	DMF

Table 2: Antibacterial activities of compounds 4(a-b) and 5(a-b)

Synthetic Compounds	Diameter of Zone of inhibition (mm)					
	Staphylococcus aureus		*Pseudomonas aeruginosa*		*Escherichia coli*	
	20 µg/ml	50 µg/ml	20 µg/ml	50 µg/ml	20 µg/ml	50 µg/ml
4a	7	8	8	9	9	9

4b	9	10	10	10	8	7
5a	-	7	-	7	10	11
5b	8	8	13	13	11	10
Amoxicillin						

Table 03: Antifungal activities of compounds 4(a-b) and 5(a-b)

Synthetic Compounds	Comp. zone of inhibition (mm)			
	Candida albiacans		*Aspergillus niger*	
	20 µg/ml	50 µg/ml	20 µg/ml	50 µg/ml
4a	9	12	10	10
4b	9	9	9	8
5a	11	13	7	8
5b	10	12	10	9
Fluconazole				

Fig.1. Scheme Of Work R: 4a-CH$_3$, 4b-C$_6$H$_5$, 5a-CH$_3$, 5b- C$_6$H$_5$

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Fig.2. IR SPECTRA
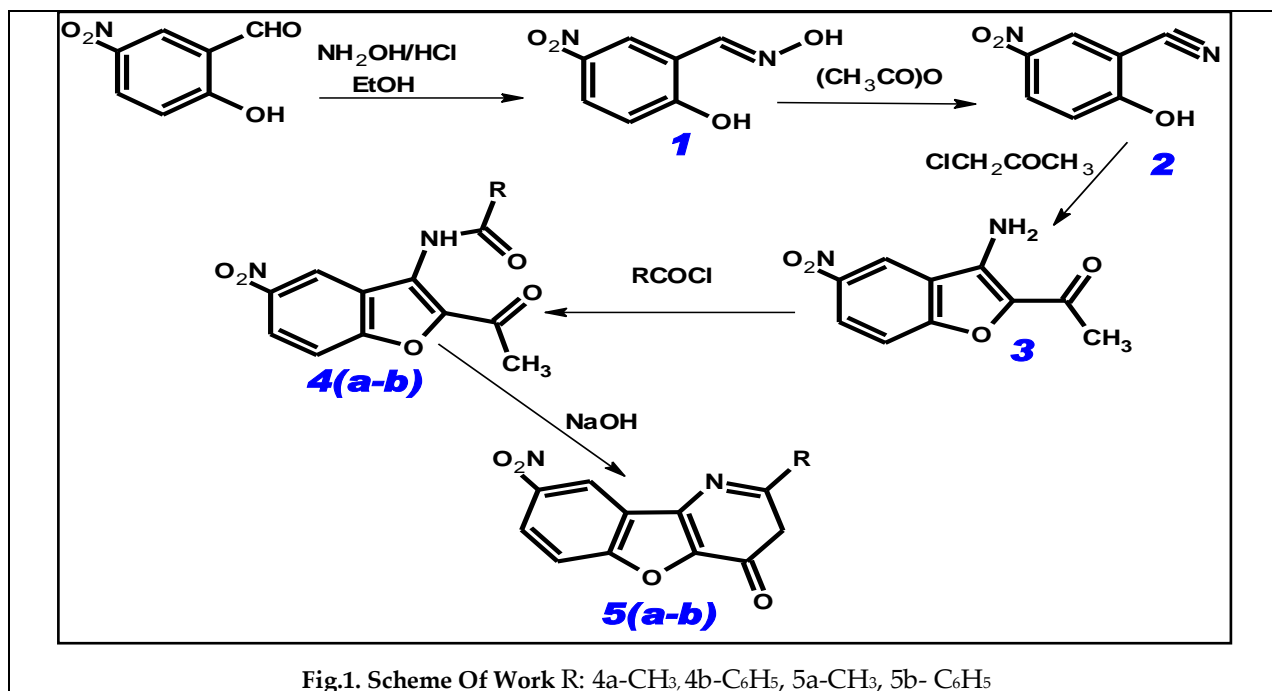
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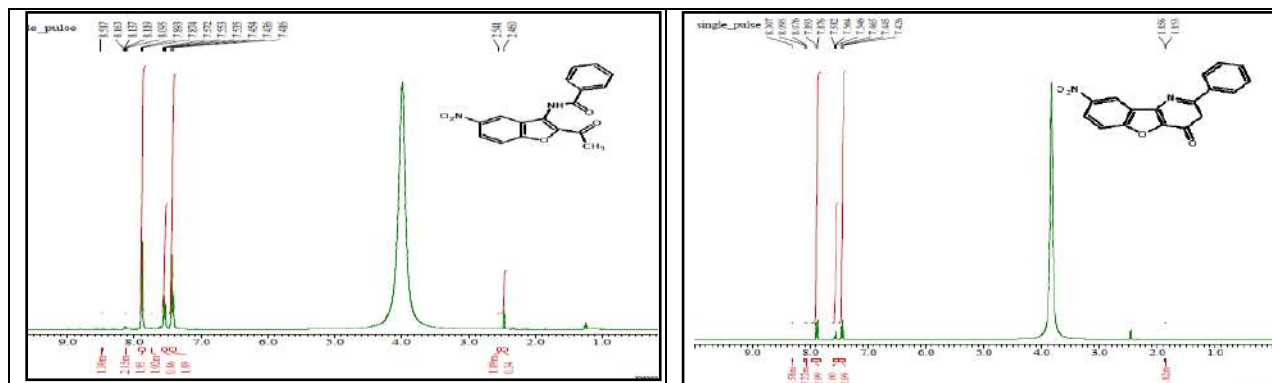
Fig.3. NMR SPECTRA
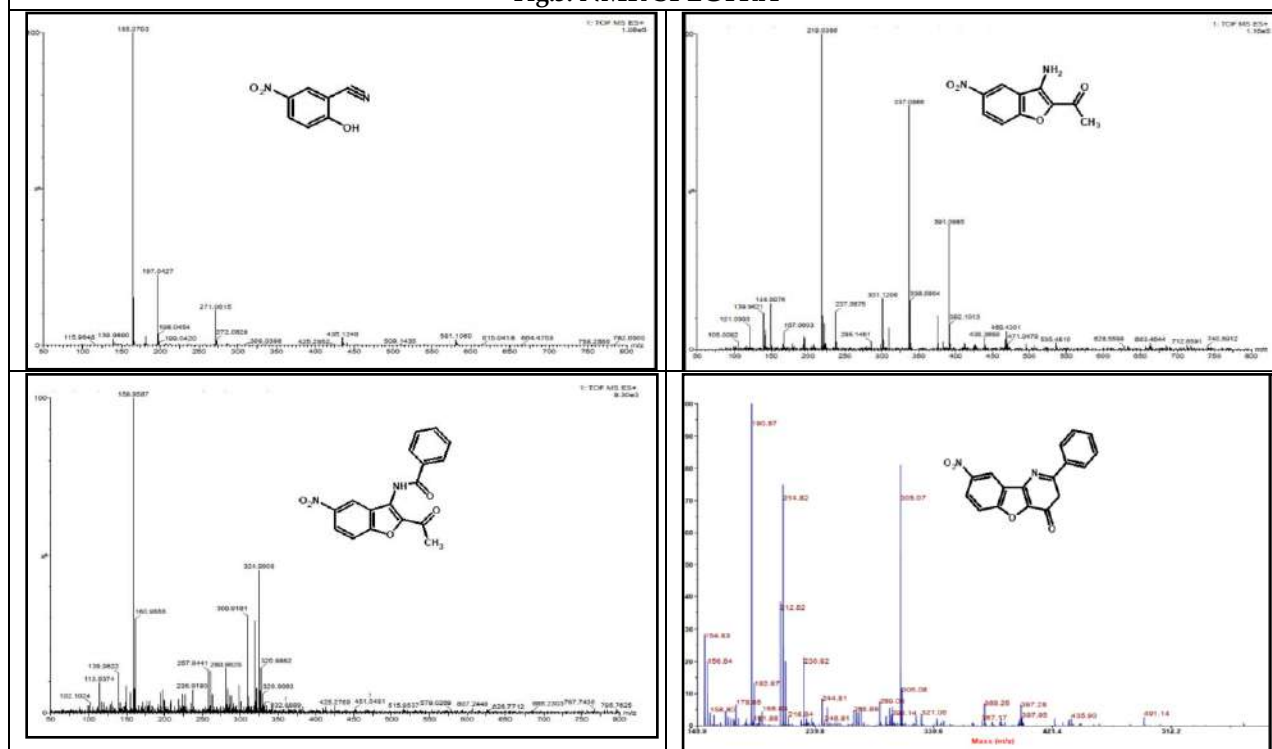
Fig.4. MASS SPECTRA
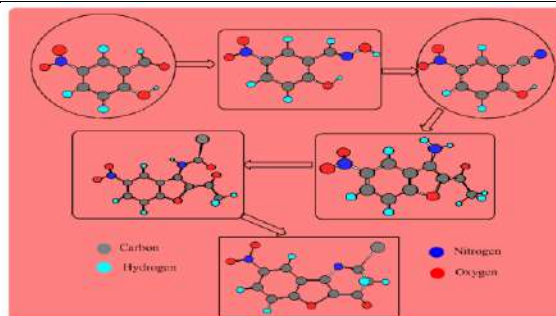
Fig.5. GRAPHICAL REPRESENTATION OF THE SCHEME

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REVIEW ARTICLE

An Insight into the Chemistry and Pharmacological Activities of Carbazole Derivatives: A Review

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ABSTRACT

Carbazole is a heterocyclic nitrogen-containing chemical which has a versatile role in different fields of research. It has a tricyclic structure with two six-membered benzene rings joined on each side with a nitrogen-containing five-membered ring (sandwiched between two 6 membered rings). Carbazomycins and murrayafoline A are two examples of naturally occurring medicinally active compounds that contain the Carbazole ring. Compounds derived from carbazole ring are biologically potent having antitumour, antibiotic, antiviral, anti-inflammatory and neuroprotective properties. The researches done in cancer chemotherapy have suggested carbazole skeleton as a promising lead for cancer treatment. The strong cytotoxic activity elicited by carbazole can be highlighted with its polycyclic and planar aromatic structure. Topoisomerase 2 inhibition exerted by carbazole derived compounds can be employed for developing novel anticancer agents which could replace the cardiotoxic top 2 poisons available in the market.

Keywords: Carbazole derivatives, Anti microbials, Anti-HIV drugs, Anti diabetics, anti convulsants, Alzheimer's disease, Cancer, TopoisomerseII ,Diarrhoea

INTRODUCTION

Carbazole is a heterocyclic nitrogen-containing chemical. It has a tricyclic structure with two six-membered benzene rings joined on each side with a nitrogen-containing five-membered ring. When exposed to ultraviolet light, it produces a powerful fluorescence and a lengthy phosphorescence [1]. Carbazole alkaloids, identified from the Rutaceae family, are a significant class of naturally occurring heterocycles. Carbazole alkaloids were first isolated as natural compounds from *Murraya koenigii* and shown to have potent antibacterial properties. *Murraya koenigii* stem bark includes dimeric carbazole alkaloids as well as six carbazole alkaloids. This herb has traditionally been used as a stimulant, stomachic, febrifuge, analgesic, and to cure diarrhoea, dysentery, and bug stings. It also has antibacterial properties in addition to these actions [2]. The isolation of ellipticine the first pyrido[4,3-b]. carbazole alkaloid, from the leaves of *Ochrosia elliptica* Labill by Goodwin et al. in 1959, was a significant milestone in the development of this class of natural compounds. The great potency of ellipticines against numerous forms of cancer (osteolytic breast cancer metastases, kidney cancer, brain tumours, and acute myeloblastic leukemia), low toxic side effects, and total lack of haematological toxicity are the key reasons for their clinical usage. Nonetheless, these chemicals' mutagenicity should be assessed as a possible risk factor for these anticancer medicines. In E. coli, they are known to produce prophage lambda. Furthermore, ellipticine has shown to be a common synthetic target, with a diverse range of synthetic techniques documented. Furthermore, the structurally similar heteroaryl annulated carbazoles have gotten a lot of attention in the lab,and these congeners have a better pharmacological profile[3].

Carbazole ring have been exploited by the research field over the past decades since the discovery of parent compound 9-H carbazole in 1972. Anti-bacterial activity of murrayanine, was the 1st discovered pharmacological activity which paved the way to further discoveries. Antiplatelet and antimicrobial activities of compounds derived from *Clausena excavata* are important among them. Other synthetic derivatives such as 5H-benzo[b]. carbazoles and furo[3,4-b]. carbazoles have proved the anti-tumour potential of carbazole derived compounds. It's possible to explore the carbazole scaffold as a solution to the challenges faced in anticancer drug research. In this review we focus on the biological potential and applications of carbazole derived compounds ranging from antimicrobial, antiviral, anti-HIV, anti-oxidant, anti-inflammatory, anti-diabetic, anticancer activities as well topoisomerase 2 inhibition in detail.

Carbazole Derivatives Exhibiting Antimicrobial Activity

Gu W et.al conducted a study on 'Synthesis and antimicrobial activities of novel 1H-dibenzo[a,c].carbazoles from dehydroabietic acid'.Through the reaction of methyl 7-oxo-dehydroabietate with a range of substituted phenylhydrazines, a number of new 1H-dibenzo[a,c].carbazole derivatives were produced in good yield. All of the chemicals were tested for their ability to kill four bacteria (*Bacillus subtilis*, *Staphylococcus aureus*, *Escherichia coli,* and *Pseudomonas fluorescens*) and three fungi (*Bacillus subtilis*, *Staphylococcus aureus, Escherichia coli*, and *Pseudomonas fluorescens*) (*Trichophyton rubrum*, *Candida albicans* and *Aspergillus niger*). Of the compounds which showed strong antibacterial properties, compound1 had antibacterial activity against *B. subtilis* that was comparable to the positive control[4]. Indolo [2,3-a]. carbazole-based inhibitors were produced from widely available indigo by Guo S et.al ina study. The inhibitors were easily and selectively functionalized at the nitrogen on the carbazole unit's indole component. The synthesised analogues have modest inhibitory activity against *Bacillus anthracis* and Mycobacterium tuberculosis, indicating that indolo [2,3-a]. carbazoles could be viable leads for the development of novel anti-anthrax and anti-tuberculosis medications[5].

The antibacterial and antifungal activities of a series of N-substituted carbazole derivatives against *Staphylococcus aureus,* methicillin-resistant *Staphylococcus aureus* (MRSA), *Bacillus subtilis, Escherichia coli, Pseudomonas aeruginosa, Bacillus proteus, Candida albicans*, and *Aspergillus fumigatus* were evaluated by Zhang FF et.al in his study 'Synthesis, antibacterial and antifungal activities of some carbazole derivatives'. Some of the synthesised compounds showed comparable or even superior antibacterial and antifungal activity than the reference medications fluconazole, chloramphenicol, and norfloxacin [6].

Antimalarial activities of amino alcohol-carbazoles were discussed by Wang T et.al in his study 'Inhibition of *Plasmodium falciparum* Hsp90 contributes to the antimalarial activities of aminoalcohol-carbazoles'. By computationally docking a broad variety of antimalarial compounds previously discovered in a phenotypic screen into a PfHsp90-specific pocket, researchers identified an amino-alcohol carbazole as a PfHsp90-selective inhibitor. Investigators performed molecular dynamics (MD) simulations in the Pf-specific drug binding pocket of PfHsp90 using the original docking poses of the ten top-ranked hits, examining their interaction and stability throughout the simulation. While the majority of the compounds became unstable and finally dissociated, compound SJ000111021 (compound2) remained in its original position and displayed significant stability inside the Pf-specific pocket. Compound2is an amino alcohol-carbazole molecule[7].

A review was conducted by Mondal A on a series of studies that have performed to synthesis a novel carbazole that will be utilised to manufacture quinoline analogue. TDR30137, a commercially available chemical that is linked to N-substituted carbazoles, has proven to be effective. In human red blood cells, an IC50 of 57 nM has been discovered. The compounds demonstrated moderate to strong antiplasmodial action against the strains. The potency of compounds with aniline substituents was found to be lower. Dihalogenated carbazole derivatives was determined to be a privileged core[8]. Zika virus sickness is caused by a virus that is mostly transmitted by Aedes mosquitoes that bite during the day. Fever, rash, conjunctivitis, muscle and joint discomfort, malaise, and headache are all common symptoms. Infection with the Zika virus (ZIKV)caused an international health emergency in the Americans in 2015-16, and despite its high visibility, there is currently no approved therapy for Zika virus infection, putting millions of people at risk. ZIKV belongs to the Flaviviridae family, which includes well-known members like dengue virus (DENV) and West Nile virus (WNV). The NS2B/NS3 protease is one of the best validated targets for developing anti-flaviviral treatments for DENV and WNV infection [9].

Four series of carbazole-based mono- and bis-amidines were synthesised in this study conducted by Rassias G et.alwith the N substituent being either methyl, benzyl, or benzenesulfonyl. Compound 3, a bis amidine derivative of N-Me carbazole, was shown to be the foremost effective compound in both the ZIKV NS3pro biochemical and ZIKV cellular infectivity assays (IC50- 0.52 M, EC50- 1.25 M) after biochemicaland cellular analysis[10]. Clausine K or clauszoline J, a natural substance isolated independently from a variety of sources, was found to have only moderate antituberculosis action (MIC of 100 mg/mL) against the H37Ra strain. Choi TA et.al tested a number of carbazole alkaloids and derivatives for anti-TB activity in vitro to see if the carbazole framework may be used as a new anti-TB scaffold. It is concluded that carbazoles could be developed as anti-TB medication candidates with further structural modification to boost anti-TB potency, compounds 4 and 5 were the most active and selective (SI>10)[11].

Anti HIV Activity

The mortal immunodeficiency contagion Human Immuno Deficiency virus attacks the vulnerable system, weakening people's defences against a variety of ails and cancers that healthy vulnerable systems can combat. The most advanced stage of HIV infection is acquired immunodeficiency syndrome (AIDS) which, depending on the existent, can take several times to develop if not treated. The development of certain tumours, infections, or other serious long- term clinical symptoms is what defines AIDS[12]. Hirata et al. synthesised N-alkyl-pyrido[4,3-c]. carbazoles from 2-hydroxy-9H-carbazole-3-carbaldehyde (mukonal), a chemical component found in Rutaceous plants, in 1999. These chemicals have been tested as antiviral agents and are structurally linked to the anticancer alkaloid Ellipticine and its synthetic analogues. Several compounds inhibited HIV replication in H9 cells, with the 5-methoxy-7-methyl-7H pyrido[4,3-c].carbazole having the highest therapeutic index (TI = 503) in the series with an EC50 of 0.0054 g/Ml[13].

The results of a preliminary biological analysis of a small series of chloro-1,4-dimethyl-9H-carbazoles for anti-HIV-1 activity are presented by Saturnino C and colleagues. The nitro-derivative compound7 had the most intriguing profile of all the compounds examined, indicating that the chlorine location on the carbazole scaffold may be

significant for its antiviral action. The findings suggest that this class of chemicals could be useful as a starting point for the creation of selective and possibly alternative methods for the treatment of HIV-related chronic illnesses[14].

Anti- Inflammatory, Anti-Oxidant Activity

Liu YP et.al extracted and identified Twelve carbazole alkaloids from the stems and leaves of *Clausena vestita*, a Chinese indigenous plant, including *Clause vestine*, a novel prenylated carbazole alkaloid, and 11 recognised prenylated carbazole alkaloids. The obtained prenylated carbazole alkaloids were evaluated for anti-inflammatory and antiproliferative properties. When compared with positive control (hydrocortisone) with IC50 values, the prenylated carbazole alkaloids had potent inhibitory effects on NO (nitric oxide) generation. Findings suggest that these prenylated carbazole alkaloids have potent anti-inflammatory and antiproliferative properties[15]. Bandgar BP et.al studied the structures of freshly synthesised 3-(substituted)-aryl-5-(9-methyl-3-carbazole)-1H-2-pyrazolines by IR, 1 H NMR, and mass spectral analysis. All of the produced compounds were tested for anti-inflammatory action in vitro and in vivo, as well as antioxidant activity. The COX-2 inhibitors were discovered to be selective. In rats, compound 8 was demonstrated to be a strong inhibitor of carrageenin-induced paw oedema. The majority of the compounds were effective at scavenging DPPH and superoxide radicals. The results of the molecular docking, together with the results of the biological assays, suggested that compound 8could be a viable anti-inflammatory drug[16].

Anti-Diabetic Activity

Eseyin OA et.al in his study 'Synthesis and in vitro antidiabetic activity of some alkyl carbazole compounds' synthesized methylcarbazole, ethylcarbazole, propylcarbazole, and butylcarbazole compounds by using acid-catalyzed alkylation method and conventional protocols were used to perform in vitro inhibitory assays on alpha amylase and alpha glucosidase enzymes on the produced compounds.. Carbazole and methylcarbazole had no inhibitory impact on this enzyme, while the reference drug (acarbose) was more effective than the produced products in inhibiting the two enzymes. Some anti-diabetic alkyl-carbazoles have been effectively synthesised[17].

Mitra Aand her co-workers demonstrated how mahanimbine (a carbazole alkaloid found in *Murraya koenigii* leaves) affects blood glucose and serum lipid profiles in diabetic rats caused by streptozotocin. The diabetic rats were given a single dose of mahanimbine (50 and 100 mg/kg) once a week for 30 days. For the same amount of time, the control group was given 0.3 percent w/v sodium carboxy methyl cellulose. In diabetic and non-diabetic rats, fasting blood sugar and serum lipid profiles were assessed. At doses of 50 and 100 mg/kg, mahanimbine lowered raised fasting blood sugar, triglycerides, low density lipoprotein, and very low-density lipoprotein levels while increasing high density lipoprotein levels in diabetic rats. When compared to acarbose, mahanimbine had a significant alpha amylase inhibitory impact and a mild alpha glucosidase inhibitory effect[18]. The antioxidant and PPARa/c dual agonist properties of a variety of hydroxycarbazole compounds were investigated by kumar R et.al. While the majority of the compounds had strong antioxidant action, several of them were also identified as possible PPARa/cdual agonists. In animal experiments, compounds3-(4-(2-(4-Methoxycarbazole)-ethoxy)-phenyl)- 2-ethoxy propionic acid, L-lysine salt and 3-{4-[2-(9H-Carbazol-4-yloxy)-ethoxy].-phenyl}- 2-(1-methyl-3-oxo-3-phenyl-propenylamino)-propionic acid, L-lysine salt were proven to be active[19].

Anti-Psychotic and Anti Convulsant Activity

Hemlata Kaur and her co-authors synthesized new carbazole derivatives by incorporating oxadiazole, thiadiazole, azetidinone and thiazolidinone pharmacophores at the 9th position of carbazole nucleus with the aim to get better antipsychotic as well as anticonvulsant drugs.among the series of compounds they have synthesized, compound 9-[40 -{2-Bromophenyl}-2-oxo-thiazolidin-100-yl-10 , 30 , 40 - thiadiazol-50 -yl]. methylene carbazole(compound 10) was found to show the promising anticonvulsant and antipsychotic activity [20].

Anti Alzheimers Activity

Alzheimer's disease (AD) is an irreversible, progressive brain ailment caused by changes in nerve cells that lead to the loss of brain cells. The activity of acetyl cholinesterase rises in Alzheimer's disease patients, leading to increased breakdown of the neurotransmitter acetylcholine and a drop in acetylcholine levels in the brain[21]. Anti-cholinesterase action has been discovered in 9H-carbazole derivatives and 2,3,4,4a-tetrahydro-1H-carbazoles. As a result, Kukreja Het.al in his study had chosentetrahydrocarbazole as the core for substitution.2,3,4,9-tetrahydro-1H-carbazole and its derivatives were created as selective acetylchol inesterase inhibitors by substituting at positions 6 and 9. Ellman's method for estimating acetyl cholinesterase and butyrylcholinesterase inhibitory activity was used to test these compounds. All of the substances inhibited both AChE and BChE. Compounds such as 6-Amino-2,3,4,9-tetrahydro-1H-carbazole, 9-Methyl-6-nitro-2,3,4,9-tetrahydro-1H-carbazole and N-Butyl-2,3,4,9-tetrahydro-1H-carbazol-6-amine were discovered to be selective AChE inhibitors[22]. Butyryl cholinesterase (BChE) inhibition could be a promising treatment target for Alzheimer's disease (AD). The anti-BChE activity of a new series of 2,3,4,9-tetrahydro-1H-carbazole derivatives was investigated by Ghobadian R *et.al*. All of the proposed compounds were found to be selective BChE inhibitors during in vitro experiments. While all of the compounds had AChE IC50 values below the detection limit (>100 M), they were effective BChE inhibitors that were selective. The most effective anti-BChE activity (IC50 value=0.11 M), best BChE selectivity, and mixed-type inhibition was found in 1-(2-(6-fuoro-1,2,3,4-tetrahydro-9H-carbazole-9-yl) ethyl) piperidin-1-ium chloride[23].

Cancer Chemotherapy

Cancer is the second biggest cause of mortality worldwide, defined by uncontrolled cell proliferation that can spread to distant regions of the body, causing serious health issues. Lung cancer (1.8 million cases, 12.6 percent of all cases), breast cancer (1.7 million cases, 11.9 percent), colorectal cancer (1.4 million cases, 9.8 percent), and prostate cancer (1.1 million cases, 7.7%) are the most commonly diagnosed malignancies globally. Given the rise in cancer incidence and the emergence of resistance to anti-cancer medication regimens, research and development of new potent cancer treatments has become critical in the coming decades. Among the extant anti-cancer medications, carbazole scaffolds have been the primary structural motif of many biologically active molecules, both natural and synthetic, for over half a century [24]. The cytotoxic activity of carbazole alkaloids from clausena excavate was investigated by Peng WW et.al. Excavatine A (1), a new carbazole alkaloid, and excavatine B (2) and excavatine C (3), two further new carbazole alkaloids, were extracted from the stems and leaves of *Clausena excavata* Burm.f (Rutaceae). Detailed spectroscopic investigations, particularly 2D-NMR and HR-EI-MS data, were used to determine their structures. Compounds 1–3 were evaluated for cytotoxicity against the cancer cell lines A549, HeLa, and BGC-823, as well as antibacterial activity against Candida albicans and Staphylococcus aureus. Only one showed cytotoxicity in A549 and HeLa cell lines, with IC50 values of 5.25 mg/ml and 1.91 mg/ml, respectively[25].

For the first time, TFAA/H3PO4 has been identified as a good coupling agent for carbazole N-acylation. This technology was used by KumaráKolli S et.al to make a number of N-acylated derivatives that were investigated for anti-proliferative effects against oral and breast cancer cell lines. Several of them were found to be promising, indicating that the current series of N-acyl carbazoles can be used to find and develop novel anticancer medicines [26]. Cell proliferation, differentiation, and death are all regulated by JAK-STAT signalling. Recent research suggests that STAT family proteins, particularly STAT3, play a vital role in promoting tumour cell transformation by specifically establishing and sustaining a pro-carcinogenic inflammatory milieu. Carbazoles have been shown to suppress STAT3-mediated transcription in a recent study. To test the activity of N-alkylcarbazole compounds substituted in positions 2, 4, and 6 on STAT3, a new set of N-alkylcarbazole derivatives was prepared by Saturnino C et.al. Some of these compounds demonstrated interesting action as STAT3 selective inhibitors[27].

Diaz P et.al investigated the effect of modified carbazoles on microtubules and glioblastoma multiform cells. Microtubules are destabilised by a novel series of modified carbazoles that bind to the colchicine site of 462 tubulins in a manner akin to a podophyllotoxin analogue and appear to interact with a unique low interaction 463 binding area. Several modified carbazoles cause cell killing in a variety of GBM model systems, but 464 has a significantly

lower activity in HepG2 liver cells, indicating a prospective therapeutic index. When compared to 212 HepG2 cells, PD-GBM cells had a higher sensitivity to modified carbazole killing activity, suggesting a possible therapeutic index (i.e., the ratio of the amount of agent that causes the 213 therapeutic effects to the amount that causes toxicity)[28]. The structure-activity correlations of novel 14 N-substituted carbazole sulfonamide derivatives with better physicochemical properties were created and examined byLiu et.al. The majority of these novel compounds had excellent water solubility. Certain compounds had potent antiproliferative and anticancer action *in vitro* and *in vivo.* In both subcutaneous and orthotopic HepG2 xenograft animal models, compound 12 showed anticancer activity [29].

Topoisomerase 2 Inhibition By Carbazole Derivatives

Topoisomerase II uses a DNA breakage/reunion mechanism to catalyse DNA topological processes. The DNA topological reactions enable the enzyme to segregate interlocked chromosomal DNA during mitosis and eliminate excess DNA supercoils created during DNA replication, RNA transcription, and chromosome condensation. Many anticancer medications, such as etoposide, anthracyclines, and m-AMSA2, can disrupt topoisomerase II's breakage/reunion mechanism, resulting in the accumulation of a topoisomerase II-DNA covalent intermediate, the cleavable complex. Tumor cells die when topoisomerase II-DNA cleavable complexes accumulate[30].. Antineoplastic medicines that inhibit topoisomerase II and induce the topoisomerase II-DNA cleavable complex are among the most effective antitumor medications now available for the treatment of human tumours, and they are important components of combination chemotherapy. However, these medications are ineffective against non-small cell lung cancer, colon cancer, gastric cancer, and pancreatic cancer, which are all prevalent solid tumours. Furthermore, most topoisomerase II inhibitors are P-glycoprotein or MRP substrates. Topoisomerase II drugs are therefore less efficient in treating P-Glycoprotein or MRPpositive malignancies. The toxicity of doxorubicin and other anthracyclines, which causes heart damage, restricts their usefulness. As a result, the researchers believed that a structurally new topoisomerase II inhibitor with greater anticancer activity in vitro and in vivo than etoposide and doxorubicin would be a strong candidate for a clinically relevant antitumor medication[31].

Novel pyrimidoacridones, pyrimidophenoxadines, and pyrimidocarbazoles were synthesised and assessed in a work done as part of a series of experiments to find powerful topo II inhibitors. The pyrimidocarbazoles were the most effective anticancer agents in vitro and in vivo. In an in vivo solid tumour model, (ER-37326) showed more powerful activity than etoposide without causing serious toxicity (M5076). Finally, the authors discovered a novel lead chemical (ER-37326) that merits further exploration as a strong anticancer treatment for solid tumours[32]. ER-37328 a novel carbazole top 2 inhibitor synthesized by Nakamura K et.al inhibited topoisomerase II activity at 10 times lower concentrations than etoposide in a relaxation assay and produced double-strand DNA cleavage in murine leukaemia P388 cells within 1 hour. At 2 M, the highest amount of DNA cleavage was achieved. In P388 cells, ER-37328 (2 M) triggered topoisomerase IIDNA cross-linking in the same way that etoposide did. ER-37328 exhibits apparent DNA binding ability, according to a spectroscopic investigation of ER-37328 combined with DNA. A panel of 21 human cancer cell lines demonstrated substantial growth-inhibitory action with ER-37328 [mean (50 percent growth-inhibitory dose) GI50 59 nM].. In vivo, ER-37328 caused significant tumour shrinkage in Colon 38 carcinoma injected s.c., outperforming etoposide and doxorubicin[33].

Saravana bhavan M et.al performed microwave irradiation in the presence of p- toluene sulfonic acid yielded a unique series of 2,3-thiophene-5,6-dihydro-11H-pyrido [2,3- a]. carbazole-4-one derivatives (3a-3g). Furthermore, compound 15 and 16 (3e and 3f) had higher cytotoxicity than the positive control Cisplatin (also compared to Ellipticine), which could be attributed to the chloro and bromo groups' increased electron withdrawing tendency. The antioxidant activity of compound 3g with a -OCH3 substitution and compound 3e with a chloro substitution is higher than that of ordinary Ascorbic acid. The proposed compound 14 and 15fit well in the active site (ATP-binding site) of the probable target (human CK2), implying that drugs may inhibit protein kinase CK2 and so have anticancer activity[34].

Carbazole amino alcohol derivatives were produced and tested as a range of new anticancer drugs in a study done by Wang W et.al.The aliphatic-amine-substituted derivatives are more powerful antitumor agents than those containing aromatic amino groups, according to SAR tests. Alkylamine-substituted compounds had the most effective antiproliferative action, which is consistent with their capacity to inhibit topoisomerase I. The representative chemicals 5–7 act as topo I poisons, causing single-strand DNA damage by stabilising the topo I–DNA cleavage complex, based on comet experiments and molecular docking investigations. Furthermore, in HeLa cells, the most potent compound 17, caused G2-phase arrest and apoptosis [35]. The effect of novel pyrrolo[2,3-a]. carbazole compounds on topoisomerase I activity in vitro, as well as the viability of glioma and endothelial cells in vitro and angiogenesis *in vivo*, was investigated by Lampropoulou E et.al. All of the studied drugs reduced the number of viable glioma and endothelium cells in vitro in a concentration-dependent manner, albeit efficiency and potency differed in endothelial cells compared to glioma cells. The most effective chemicals in glioma cells and the most efficient compounds in decreasing the number of viable endothelium cells were 1d2(compound18) and 1e (compound 19).Finally, all of the compounds examined blocked angiogenesis in the chorioallantoic membrane of chicken embryos in a considerable and dose-dependent way, with compound 1d2(compound 18) being the most effective inhibitor. These findings show that the pyrrolo[2,3-a]. carbazole compounds examined decrease topoisomerase I activity and may be effective in glioma cell proliferation and angiogenesis inhibition[36].

Pyrido [3,2]. carbazide derivatives are found to be effective in the A549 cells of lung cancer and HT29 cells of colon cancer therapy. Knoevenagel condensation and intramolecular Heck-type reactions are used to successfully synthesis the intermediates resulting in a unique and efficient synthetic method to the target compounds' core scaffold. With IC50 values ranging from 0.07 M to 4.45 M, these target compounds showed an intriguing anticancer profile toward the examined cell lines. The best compound (compound 20), against A549 and HT29 cells, is 7 times and nearly 10 times as powerful as R16[37]. A variety of carbazole derivatives including chalcone analogues (CDCAs) were produced and their Topo II inhibition and cytotoxic effects were explored in research done byLi PH et.al. The results of the Topo II-mediated DNA relaxation assay revealed that CDCAs can considerably reduce Topo II activity, and the structure-activity connection revealed that the halogen substituent in the phenyl ring is critical in the activity. CDCAs are nonintercalative Topo II catalytic inhibitors, according to additional mechanistic investigations. Furthermore, certain CDCAs exhibited micromolar cytotoxicity. The most effective chemical, 3h (compound 21), inhibited the development of four human cancer cell lines significantly. Compound 3h promotes apoptosis through the activation of caspase proteins, according to Western blot research [38].

WenGu et.al studied on 'Synthesis, in vitro antimicrobial and cytotoxic activities of new carbazole derivatives of ursolic acid'. All the synthesized compounds were evaluated for their antimicrobial activity against four bacterial and three fungal strains using serial dilution method. Potent antibacterial activity against the tested bacteria was showed by compound 22with 5-fluoroindole moiety and N-(dimethylamino)propyl amide side chain[39].

Neurological Activity

The most common neurologic infection is epilepsy, which is characterised by excessive transient neuronal discharge. The illness affects up to 50 million individuals globally and affects 1.0 percent of the population. Previous research has shown that a considerable percentage of people taking anti-epileptic medicines (20%–30%) are resistant to the present treatment agent[40]. Rajamanickam et al. presented the N-substituted carbazoles 82–87, which were tested for antilepileptic and antinociceptive properties. At a dosage of 20 mg/kg, compound 86 demonstrated significantly considerable anti-epileptic potential. This might be owing to the substituent 2-(2, -3-dimethylphenyl)-amino-benzoic acid, which was published in Molecules in 2015. Analgesic properties have been found for compounds 82–87[41].

Neuro2a cells are also commonly employed to research neurite development and neuronal differentiation. Neurite outgrowth is recognised to be vital for neuronal plasticity and regeneration and these functions are important for designing neuronal regeneration therapies in the case of nerve injury and neurological diseases. Research done by Furukawa Y et.al. discovered that, one of the 22 carbazole compounds, compound 3-ethoxy-1-hydroxy-8-methoxy-2-methylcarbazole-5-carbaldehyde, may protect neuro2a cells from hydrogen peroxide-induced cell death. Neurite loss

is one of the most well-known characteristics of neuronal damage. Compound was found to be able to trigger neurite outgrowth in neuro2a cells via the PI3K/Akt signalling pathway in the current investigation. These data imply that compound 23may have a neurotrophic impact, making it a potential treatment for brain injury[42].

Anti-Emetic Activity

Vomiting happens when the emetic centre in the medulla oblangata is stimulated. Antiemetics are medications that are designed to prevent or control vomiting[43]. Ondansetron is a carbazole derivative that works as a competitive serotonin type 3 receptor antagonist and is used to relieve nausea and vomiting produced by cytotoxic chemotherapy drugs. Ondansetron is a selective antagonist of the serotonin 5-HT3 receptor. The drug's antiemetic effect is mediated via the inhibition of 5-HT3 receptors located both centrally (medullary chemorecemptor zone) and peripherally (peripheral chemoreceptor zone) (GI tract). This suppression of 5-HT3 receptors decreases the vomiting center's visceral afferent stimulation, both indirectly at the level of the area postrema and directly at the level of serotonin activity inside the area postrema and the chemoreceptor trigger zone[44].

Anti-Diarrhoeal Activity

Diarrhoea is defined by an increase in the water content, volume, or frequency of stools, as well as a disturbance in regular bowel movement. Inflammatory mediators (prostaglandins, leukotrienes, bradykinin, nitric oxide), hormones, neuropeptides, intestinal wall integrity, circulatory system efficiency, and enteric nervous system efficiency all play a role in intestinal water balance [45]. Mandal S et.al performed the separation of three bioactive carbazole alkaloids, kurryam (I), koenimbine (II), and koenine (III), from the n-hexane extract of *Murraya koenigii* Spreng's (Rutaceae) by a bioassay. The compounds' structures were verified using 1H, 13C, and 2D NMR spectrum data. Castor oil-induced diarrhoea and PGE2-induced enteropooling in rats were significantly inhibited by (I) and (II) of the three compounds. In the charcoal meal test, the chemicals caused a substantial decrease in gastrointestinal motility in Wister rats[46].

The possible non-competitive antagonists of the GluK2 receptor, indole and carbazole derivatives, were produced and studied. The position of the binding site in the receptor transduction domain inside one receptor subunit is compatible with molecular docking and molecular dynamics of ligand-receptor complexes. The produced chemicals might be used to treat disorders involving imbalanced glutamatergic transmission, such as epilepsy and neurodegenerative diseases (Parkinson's disease, Alzheimer's disease, and Huntington's disease). It's worth noting that the presumed non-competitive receptor blocking technique is good for the medication profile and may lessen adverse effects[47]. To produce 2,3-dialkylindoles and carbazoles, Fischer indolization was easily accomplished using samarium iodide, an in-silico docking analysis was performed on all novel drugs which bind the MDM2 receptor and Peripheral benzodiazepine receptor (PBR) proteins. For all 2,3-dialkylindoles and carbazoles, structures were determined using 1H NMR, 13C NMR, and LCMS. The 1,3-difluoro-6,7,8,9-tetrahydro-5H-carbazole and 1,3-dichloro-6-neopentyl-6,7,8,9-tetrahydro-5H-carbazole in PBR protein showed selected good interaction with active site amino acids such as GLN20, LYS21 with binding energy of -2.494 kcal/mol[48].

To further understand the nature of 5,6-dihydro 11-alkylbenzo[alpha]. carbazole derivatives and examine the interactions of homolog series with binding sites on chosen a-chains of human oestrogen receptors (hER) QSAR and docking experiments were used. The best QSAR model was chosen, with a r = 0.924 correlation coefficient, r2 = 0.854 squared correlation coefficient, s = 0.357 standard deviation, and Q2 = 0.755 cross-validated squared correlation coefficient. The descriptors E-HOMO and heat of formation have a crucial influence in human oestrogen receptor inhibitor actions, according to the QSAR model. The interactions between the selected compounds as oestrogen inhibitors and the human oestrogen receptor were visualised using docking research[49].

CONCLUSION

From the above topics covered we could conclude that the carbazole moiety has an important role in the pharmaceutical field. Most of the drugs now we use for various chronic conditions are derived from carbazole heterocycle. Carbazole and its derivatives are essential nitrogen-containing heterocyclic molecules found throughout nature. Because of their vast applicability, carbazole nuclei have been the subject of several research projects. Carbazole derivatives show promise as a therapeutic candidate for a variety of disorders. Carbazoles have anti-cancer, antibacterial, antifungal, anti-inflammatory, anti-histaminic, antioxidant, hepatoprotective, anti-HIV, antiprotozoal, anti-tubercular, anti-epileptic, and sedative characteristics, as well as the capacity to block topoisomerase II. Many species, such as bacteria, fungi, and parasites, are resistant to carbazole derivatives. Carbazole derivatives have the potential to be multifunctional therapeutic agents for neurological diseases. Also, *in-silico* studies were performed to elucidate the biological potential of carbazole derived compounds. Novel topoisomerase 2 targeting antitumour agents can be produced by some modifications of carbazole ring, as the studies have already proved the top 2 inhibiting activity of carbazole derivatives.

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Carbazole ring | Compound1 | Compound2

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Compound 3

Compound 4

compound5

clausine K
(clauszoline J)

Compound6

Compound 7

Compound 8

Compound 9 (mahanimbine)

Compound 10

(Compound 11) Excavatine

Compound12

Compound 13(ER 37326)

Compound14(ER37328)
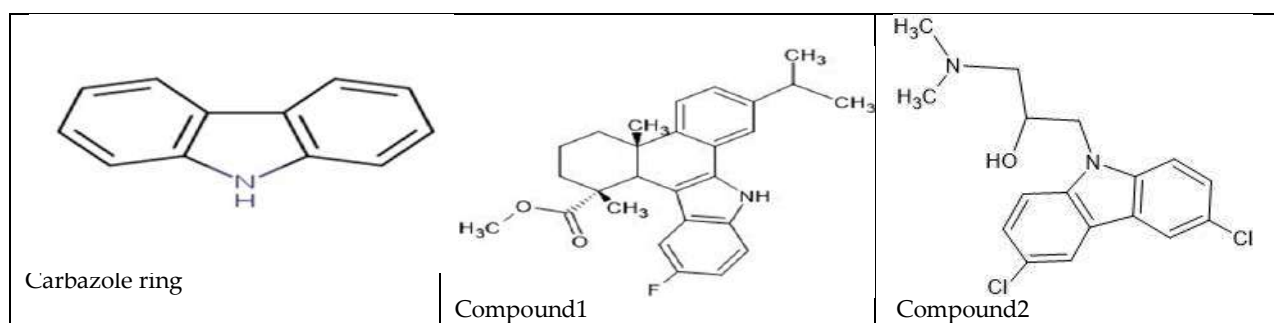
Compound 15
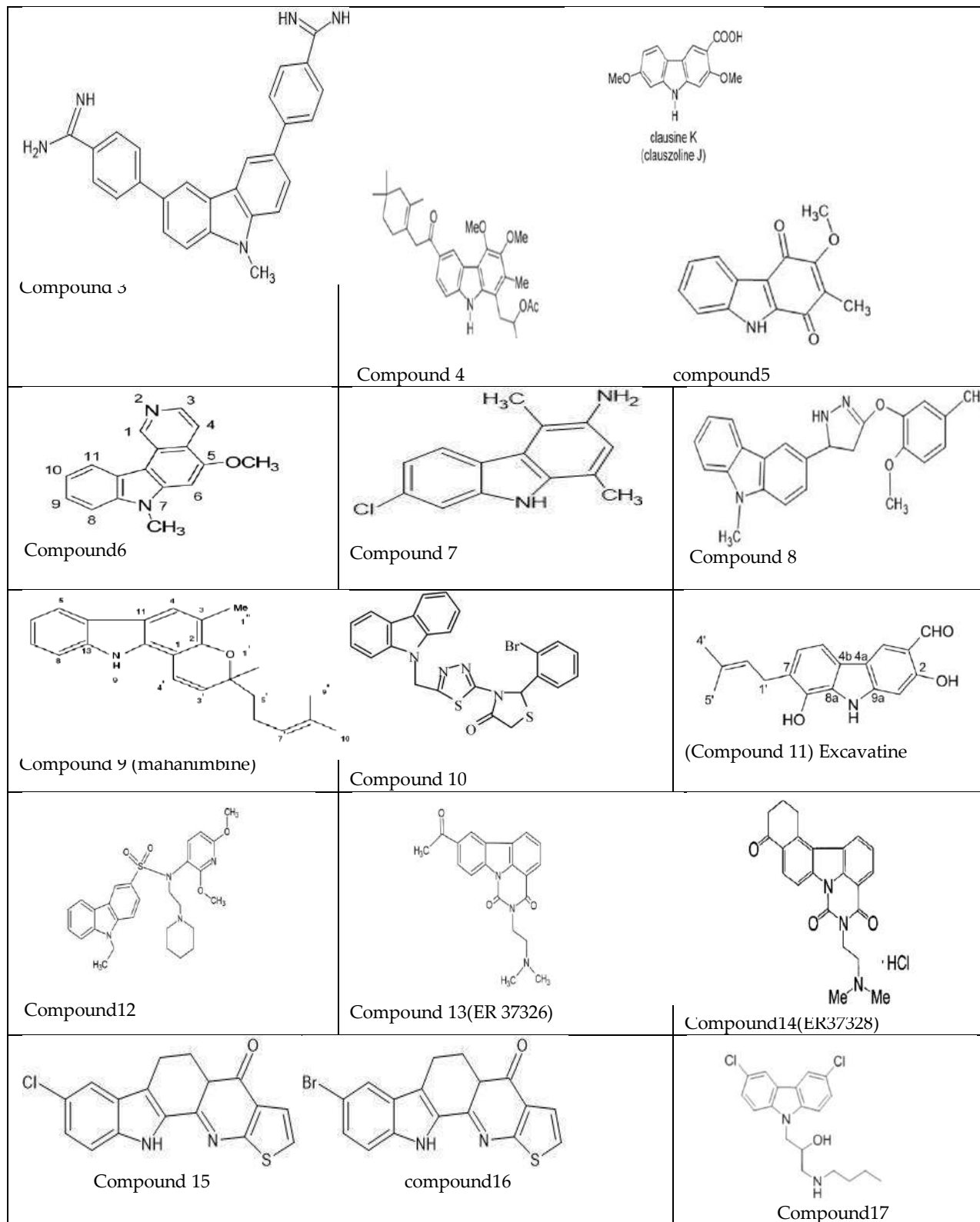
compound16

Compound17

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Compound 18 compound19		Compound 20
Compound 21	Compound 22	Compound 23
Compound 24(ondansetron)	Compound 25(1,3-dichloro-6-neopentyl-6,7,8,9-tetrahydro-5H-carbazole)	

RESEARCH ARTICLE

Protective Effect of *Cissus quadrangularis* on Carbosulfan Induced Nephrotoxicity in Male Albino Rats

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ABSTRACT

Several insecticides are used in agriculture industry to protect the crops; in contrast the farmers are continuously being exposed to these insecticides and suffer from induced insecticidal toxicities over the time. The present investigation was carried out to evaluate the ameliorative effects of *Cissus quadrangularis* (CQ) plant extract on carbosulfan induced toxicities in male albino rats. The rats were divided into 6 groups; Control 1% DMSO (K_1), Plant extract 1000mg/kg bw (K_2), Low dose carbosulfan 3.4 mg/ kg bw (K_3), High dose carbosulfan 6.37 mg/ kg bw (K_4), plant + low dose carbosulfan 1000 mg + 3.4 mg/ kg bw (K_5), and plant + high dose carbosulfan 1000 mg + 6.37 mg/ kg bw (K_6). The treatment was given intragastric via oral gavage for 60 days. Histopathological studies of experimental group rat kidney exhibited various pathological lesions including hemorrhage, pyknotic nuclei, dilated Bowman's capsule, altered glomerular structure, and shrunken glomeruli. The lipid profile and hormonal variations were also significantly altered in carbosulfan treated groups and indicated hyperlipidemic and induced hypothyroidism effects. However, the protective treatment of CQ extract significantly recovered the lipid profile, hormone level and histopathological alterations. The high dose carbosulfan treated group was affected more. While, at the same time the treatment of CQ manifested mild recovery rate in this group (K_6). The results suggest that the plant extract was able to recover the damages caused by the carbosulfan treatment. These results provide the crucial data required for future ameliorative drug development purposes.

Keywords: *Cissus quadrangularis,* Carbosulfan, Kidney, Histopathology, Thyroid hormone

INTRODUCTION

In recent years, the agricultural sector has revolutionized with the involvement of new technology including high crop-producing plants, storage, and pest control methods. The majority of farmers still encounter crop-damaging pest species that are destructive in nature. Control of pests in the agriculture field is one of the most important things to manage to get a high crop yield. The synthesis of organophosphate compound (OPC) in 1848 initiated the era of pesticide formulations [1]. Since then, the new combination of pesticides was synthesized and introduced into the market for use. However, toxicity studies of organophosphates on organisms including fishes, rodents, and amphibians indicated the devastating effects on non-target organisms [2], [3]. Carbamates were introduced during the mid-19th century to overcome the high toxicity of organophosphates [4]. Carbosulfan [2,3-dihydro-2,2-dimethyl-7-benzofuranyl [(dibutylamino)thio] methylcarbamate is one of the carbamate derivatives that is extensively used as an insecticide in Most part of the world. It is effective against a wide range of pests such as green leafhopper, caterpillars, brown planthopper, gall midge, stem borer and leaf folder of paddy, white-backed planthopper, white aphids of chilies and it was also effective on pyrethroid-resistant mosquitoes [5]. However, In abiotic conditions (absence of organic solvents & minerals), it can persist longer period in natural water [6]. Recent statistical data released by the ministry of agriculture & farmers welfare, Govt of India, revealed that Karnataka was used 1608 MT (metric tonnes) of chemical pesticides for agriculture purposes and India was found to utilize 28 MT of technical grade carbosulfan in 2019-20 [7]. Initial screening of insecticides for their toxicity is the most important laboratory procedure to protect future intoxication of non-target organisms in nature.

Several studies were conducted on aquatic & terrestrial (non-target) organisms to screen toxic effects of the carbosulfan owing to its persistence in nature. For example, different concentrations of carbosulfan exposure induced oxygen deprivation and behavioural anomalies in *Cyprinus carpio* [8], whereas it was mutagenic & genotoxic in *Channa punctatus* and in the rainbow trout *Oncorhynchus mykiss* [9]. In *Labeo rohita*, it was found to induce oxidative stress via the generation of free radicals and altering the status of antioxidant enzymes [10]. In mammalian models, carbosulfan affects various physiological functions that are crucial for living an organism. Oral treatment of carbosulfan altered the biochemical profile and the histology of the liver in both male and female mice [11]. Exposure of carbosulfan to pregnant dams caused developmental disabilities in the offspring, indicating neuronal damage & embryonic propensity of the insecticide [12]. In an *in vivo* study, it was found to cause cell cycle delay by inducing sister chromatid exchange in the bone marrow cells of the mice [13].

Another study revealed altered enzymes, antioxidants, biochemical, and haematological responses in rats upon exposure to carbosulfan [14]. Farmers are the prime victims of occupational health hazards caused by pesticide exposure owing to the cost of protection kits, pesticides, and poor integrated pest management systems. A recent study elucidated the mean protection and health cost of pesticide exposure per farmer per vegetable season in 2019 was US $3.60 in Pakistan [15]. An *In vitro* experiment conducted on cultured human lymphocytes treated with carbosulfan resulted in chromosomal aberrations indicating the gene level of mammalian toxicity [16]. Accidental or purposeful pesticide self-poisoning or suicidal cases in rural India indicates the impact of pesticides on human beings [17].

It is necessary to first aid the pesticide-exposed patient with available home remedies before hospitalization. The traditional medicine method is one of the oldest and quick remedies to treat such pesticide cases. Plant extract-mediated amelioration of pesticide-induced toxicities was found to be promising and well documented in recent years [18]. The hepatoprotective potential was noticed in the pesticide intoxicated rats upon treatment of chicory & artichoke leaves [19], and *Citrus* lemon fruit extracts in rats [20]. *Cissus quadrangularis* (CQ) is an indigenous medicinal plant of India and belongs to the family Vitaceae. The plant has been used as an anti-dyspeptic, analgesic, anthelmintic, digestive tonic, and bone fracture healing medicine. The plant possesses strong antioxidant and antimicrobial property that serves several application in the medicinal world [21].

Previous studies explored the anti-inflammatory, anti-osteoporotic, and venotonic effects of CQ in rat models [22], [23]. The plant extract was shown to be non-toxic and exhibited a no-observed-adverse-effect-level (NOAEL) dose of 2.5 to 3g/kg bw (bodyweight)/ day in rats [24]. Considering the practical exposure of farmers to the pesticides, there is a need to develop ameliorative drugs which can bring down the pesticide toxicity. In this context, the present investigation was aimed to evaluate the protective effect of *Cissus quadrangularis* extract on carbosulfan (a carbamate derivative) induced renal toxicity in male rats.

MATERIAL AND METHODS

Collection and preparation of plant extract
The fresh stem of *Cissus quadrangularis* was collected from the surroundings of Nelamangala taluk, Bengaluru rural District, Karnataka State, India (13°3'43"N & 77°20'32"E). The stem was washed twice in tap water then soaked in distilled water. The material was shade dried for 30 days, powdered, and sewed to get the final dry product of the plant. The powder was then subjected to 10 hours of soxhlet apparatus by using ethanol & distilled water (75ml: 25ml v/v) as a solvent to extract the biomolecules present in it. A rotary evaporator (BUCHI rotavapor R-210) was used to get the final solvent-free ethanoic extract that was used for the present study.

Chemicals
Carbosulfan (Marshall 25% EC, 27.18g of active ingredient) was purchased from FMC corporation India Pvt, Ltd. Mumbai, Maharashtra. Benzene, Eosin (water-soluble), Xylene, paraffin wax, and all other chemicals were analytical grade & purchased from SRL Pvt. Ltd.

Animal Maintenance
For the present study, thirty-six adult male albino rats of Wistar strain (*Rattus* norvegicus) weighing 200-250g (7 to 8-week-old) were obtained from the animal house facility, Department of Zoology, Karnatak University, Dharwad (No. 693/GO/Re/S/02/CPCSEA). Animals were maintained in polypropylene cages with access to feed (VRK nutritional solutions, Sangli, Maharashtra) and water *ad libitum*. The rats were allowed to acclimatize to a laboratory environment for 10 days before the experimentation. The laboratory was maintained at a temperature of 26 ± 2 °C, 12/12h day & night cycle, and relative humidity of 55 ± 7% during the study period.

Experimental Design
In the present investigation, dose fixation was made based on the no-observed-adverse-effect-level (NOAEL) as depicted in the present study following OECD guidelines [25]. The selected doses were administered daily for 60 days during daylight. The animals were randomly selected and divided into 6 groups containing 6 animals in each group (n=6). The first group K_1 was the control group with the administration of 1% DMSO/ kg bw, the second group K_2 was dosed with plant extract only (1000 mg/ kg bw), third group K_3 received a low dose (3.4 mg/ kg bw) and fourth group K_4 received a high dose (6.37 mg/ kg bw) of carbosulfan, fifth group K_5 administered with plant extract plus carbosulfan low dose (1000 mg + 3.4 mg/ kg bw) and sixth group K_6 received plant extract and carbosulfan high dose (1000 mg + 6.37 mg/ kg bw).

Histology
The rats were sacrificed (inhalation of diethyl ether) on completion of the 60th experimental day. The kidneys were dissected out & fixed in formalin, then the organs were subjected to a dehydration process with different gradients of alcohol and embedded in paraffin wax [26]. A longitudinal section of (5μm thin) kidney was taken using an automated microtome (Leica RM 2255). The sections were then stained with Hematoxylin & Eosin and photographed using a Magnus phase-contrast microscope (Magnus MLXi plus, India) with an attached photographic camera (MagCam DC 5).

Hormone assay and Lipid profile tests

For the hormone assay, the blood was collected by cardiac puncture and was subjected to centrifugation at 3000 rpm for 10 minutes. Then, the serum was separated and stored in a deep freezer (-40 ℃). The hormonal assay was performed in a fully automated analyzer (Cobas e411) using the electrochemiluminescence immunoassay (ECLIA) method. The serum was analyzed to determine the concentrations of Thyroid Stimulating Hormone (TSH) and lipid profile tests such as total cholesterol, triglycerides, high-density lipoprotein (HDL), and low-density lipoprotein (LDL). Uric acid levels were also examined as an indicator of kidney functionality.

Ethical Approval

In the present study, all experiments were performed by following the guidelines of the Institutional Animal Ethics Committee (IAEC) (No. 693/GO/Re/S/02/CPCSEA), Karnatak University, Dharwad. The experimental animals used in the study were handled with care according to the guidelines provided by the Committee for the Purpose of Control and Supervision of Experiments on Animals (CPCSEA), New Delhi, India. No animals were mishandled during the experimental period.

Statistical Analyses

All the statistical data represented in the study were carried out using Origin statistical software package ver. 2019b. The data were subjected to One Way ANOVA (ANalysisOfVArience) with Tukey's post hoc test. Results were reported as mean plus or minus standard deviation ($x \pm SD$) and $p < 0.05$ was considered statistically significant.

RESULTS

Histology

The histological examination of the kidneys section of control (K_1) and plant extract (K_2) treated group manifested normal histoarchitecture. The Bowman's capsules were covered by an outer layer of simple squamous epithelium and an inner wall with flattened epithelial cells. The glomerulus was surrounded by a cluster of capillaries. Distal convoluted tubules (DCT) were numerous with low columnar cells and a central lumen. Proximal convoluted tubules (PCT) were round and consisting of pyramidal cells with a well-organized structure. The low dose carbosulfan treated group (K_3) kidney sections showed varied degrees of histoarchitectural changes (Figure 1). Dilated Bowman's capsules were the most commonly seen pathological change in this group. In addition to it, the K_3 group was also found with mild hemorrhage, altered glomerular structure, and few pyknotic nuclei. The high dose carbosulfan treated group (K_4) kidney sections were found with pyknotic nuclei, hemorrhage, increased alteration in glomerular structure, and shrunken glomeruli leading to increased Bowman's capsular area. The glomeruli structures were severely altered compare to the low dose group (K_3). In-plant extract plus low dose carbosulfan treated group (K_5) the kidney structure was shown mild alterations compare to low dose treated group (K_3). However, glomerular atrophy, hemorrhage, and alteration in the glomerular structure were frequently noticed. The recovery was found in the Bowman's capsule, where the capsular area was not altered and appeared normal. The plant extract plus high dose carbosulfan dose treated group (K_6) kidney sections showed mild recovery, but pathological changes such as shrunken glomeruli, dilated Bowman's capsule, and hemorrhage was observed with mild recovery rates.

Fig. 1: Kidney sections of post experimental rat exposed to carbosulfan and *Cissus quadrangularis*. [(K_1) Kidney section of control rat: (G) Glomerulus; (BC) Bowman's capsule; (PCT) Proximal convoluted tubule; (DCT) Distal convoluted tubule. H&E. (K_2) Kidney section of plant extract treated rat: (G) Glomerulus; (BC) Bowman's capsule; (PCT) Proximal convoluted tubule; (DCT) Distal convoluted tubule. H&E. (K_3) Kidney section of carbosulfan low dose treated rat: (H) Hemorrhage; (PN) Pyknotic nuclei; (DBS) Dilated bowman's space; (AGS) Altered glomerular structure. H&E. (K_4) Kidney section of carbosulfan high dose treated rat: (H) Hemorrhage; (PN) Pyknotic nuclei; (SG) Shrunken glomerulus. H&E. (K_5) Kidney section of plant + carbosulfan low dose treated rat: (GA) Glomerular atrophy; (H) Hemorrhage; (AGS) Altered glomerular structure. H&E. (K_6) Kidney section of plant + carbosulfan high

dose treated rat: (HGC) Hypertrophy of glomerular chamber; (H) Hemorrhage; (DBS) Dilated bowman's space. H&E.

Hormone assay and Lipid profile tests

In the present study Thyroid-stimulating hormone (TSH) was examined and represented in table 1. The control and plant extract-treated groups were found with normal levels of TSH. In the carbosulfan low dose treated group (K_3) a non-significant decrease (4.08 ± 0.69) in the hormonal levels was observed. The high dose treated group (K_4) manifested a significantly decreased level of hormone (3.11 ± 1.15) compared to control groups. The low dose carbosulfan group co-treated with plant extract (K_5) recovered the altered level of hormone but the plant plus high dose treated group (K_6) showed mild recovery in the hormonal levels (3.75 ± 0.86 SD). The uric acid profile of the control and plant extract-treated groups showed normal levels. The carbosulfan low (K_3) and high dose (K_4) treated group manifested a significant decrease in uric acid levels with 5.16 ± 0.36 SD & 4.1 ± 0.26 SD respectively. The K_5 and K_6 group which were treated with plant extract shown recovered levels of uric acid. The K_6 group was found with significant recovery upon treatment of plant extract. The lipid profile test was conducted and the results were tabulated in Table 1.

The total cholesterol levels were altered in low and high-dose carbosulfan treated groups with 213.16 ± 5.91 SD & 241.83 ± 15.17 SD respectively. Upon co-treatment of plant extract the cholesterol levels were significantly recovered compare to control groups. The plant + low dose carbosulfan treated group showed high recovery rates in the cholesterol levels. Triglycerides were significantly declined in K_3 (132.81 ± 1.74) and K_4 (117.88 ± 1.64) groups whereas in K_5 (148.35 ± 2.07) & K_6 (136.2 ± 2.77) groups it was significantly recovered compared to K_3&K_4 groups. The control (K_1) and plant extract treated (K_2) groups shown insignificant and normal levels of high-density lipoproteins (HDL). The carbosulfan low and high-dose treated groups were found with elevated levels of HDL. The elevated levels were recovered upon plant extract treatment in K_5& K_6 groups. The low-density lipoproteins (LDL) were elevated in K_3 and K_4 groups compared to the control groups. However, the treatment of plant extract recovered the altered levels of LDL in the K_5& K_6 groups.

DISCUSSIONS

Accidental pesticide exposure is one of the major issues in biological studies. The majority of farmers expose themselves knowingly or unknowingly to the dangerous combinations of drugs called pesticides. This situation has stimulated scientists to study the effects of pesticides on human beings under the domain of occupational hazards and human health. Preclinical evaluation of the drug plays a key role in the pharmacy world, however, at the same time research on first aid therapies or medicines for the accidental exposure of pesticide is important to save the intoxicated person. Histological examination of vital organs provides the best insight into the health condition of an organism. In the present study, the kidney was evaluated for the histopathological alterations induced by the carbosulfan and the ameliorative effect of the plant extract (*Cissus quadrangularis*). The kidney is the crucial target organ for several xenobiotic compounds that produces a variety of renal toxicities including the destruction of renal tubule and glomeruli. The kidney sections of experimental groups revealed several histopathological changes such as hemorrhage, pyknotic nuclei, dilated Bowman's capsule, altered glomerular structure, and shrunken glomeruli. Frequent occurrence of pyknotic nuclei in the high-dose carbosulfan treated group might be the indication of cellular toxicity in the kidney. Previous studies observed similar pathological lesions in the kidney of rats that were exposed to cisplatin. However, these induced lesions were recovered upon treatment of zingerone [27].

Glomerulus is the functional unit of kidney that receives the blood and filters the toxic substances present in it. The increase in the concentration of toxic substances impacts on the histology of the glomerulus that can be ascertained through histological changes including dilated Bowman's capsule and shrunken glomerulus. The rats exposed to organophosphates resulted similar pathological observations with glomerular atrophy and necrosis [28]. Non-permanent lesions in the kidney are often recoverable upon medicinal treatment, in our study the kidneys of rats

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exposed to carbosulfan were recovered upon treatment of CQ extract. However, the high dose treated group of rats posed mild recovery rates due to the major impact of carbosulfan. In human the kidney-thyroid axis was well studied in the medical & kidney transplantation field. Previous study reported inverse co-relation between the kidney and the thyroid axis in humans [29].

In the present study carbosulfan treatment decreased the TSH levels and upon plant extract treatment it was recovered, but complete recovery was not attained. This suggests that the long term carbosulfan treatment might induce hypothyroidism in rats. Similar alterations in the TSH levels were reported in previous studies [30], [31]. Uric acid is one of the renal injury markers in rats. During the renal filtration the uric acid generated and excreted out of the body. Renal failure due to toxicant exposure might induce accumulation of uric acid in the kidney affecting uric acid reabsorption in nephron proximal tubules. In our study the histopathological examination revealed damaged Bowman's capsule and atrophy, this supports the decreased uric acid levels owing to histoarchitectural changes in the kidney.

Recent study also found similar altered uric acid levels in the rats exposed to different concentrations of pesticide [32]. Another study suggested and considered the uric acid levels as an important factor related to kidney dysfunction [33]. The current study was also found that the lipid profile of the carbosulfan treated groups drastically increased. However, upon plant extract treatment it was recovered. The increase in lipid profile indicates the hyperlipidemic property of the insecticide. Carbamates were previously has been known to induce hyperlipidemic effects in rats. The lower level of lipoprotein concentration might be due to disruption in the formation of lipoprotein in the rat. The present results are in concordant with previous studies that worked on carbamate pesticides and obtained similar kind of effects [34], [35].

CONCLUSION

From the results obtained in the present investigation, it is evident that the *Cissus quadrangularis* plant extract was able to bring down or ameliorate the toxicity caused by the insecticide carbosulfan. Histopathological alterations in the kidney including hemorrhage, pyknotic nuclei, dilated Bowman's capsule, altered glomerular structure, and shrunken glomeruli indicates the harmful effect of the carbosulfan. However, the histopathological alterations were recovered upon the treatment of plant extract. Although, some of the pathological lesions were occurred frequently, most of the pathological changes were recovered. The lipid profile of the rat was also affected by the insecticide exposure, this could be due to the hyperlipidemic effect of the insecticide. The hormonal variation also suggests the hazardous nature of insecticide. However, the plant extract significantly recovers the physiological hormone and lipid profile levels, thereby indicating the protective effect of *Cissus quadrangularis*.

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CONFLICT OF INTEREST
Authors declare no conflict of interest.

AUTHOR CONTRIBUTION
All authors have equal contribution in the present study.

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Table 1: Lipid profile and hormonal changes influenced by the treatment of carbosulfan and *Cissus quadrangularis* plant extract in male albino rats

Parameters	Control (K_1)	Plant alone (K_2)	Low dose of carbosulfan (K_3)	High dose of carbosulfan (K_4)	Plant + Low dose carbosulfan (K_5)	Plant + High dose carbosulfan (K_6)
TSH level ($mIUL^{-1}$)	5.31 ± 1.34 SD	5.4 ± 0.63 SD	4.08 ± 0.69 SD	3.11 ± 1.15* SD	4.71 ± 1.02 SD	3.75 ± 0.86 SD
Uric acid ($mgdL^{-1}$)	6.28 ± 0.51 SD	6.35 ± 0.28 SD	5.16 ± 0.36* SD	4.1 ± 0.26* SD	5.73 ± 0.33 SD	4.98 ± 0.23* SD
Total cholesterol ($mgdL^{-1}$)	208.3 ± 2.16 SD	186.68 ± 2.49 SD	213.16 ± 5.91 SD	241.83 ± 15.17* SD	198 ± 7.15 SD	224.83 ± 20.42 SD
Triglycerides ($mgdL^{-1}$)	153.1 ± 1.70 SD	158.15 ± 1.46 SD	132.81 ± 1.74* SD	117.88 ± 1.64* SD	148.35 ± 2.07* SD	136.2 ± 2.77* SD
High-density lipoproteins	46.6 ± 4.28 SD	50.3 ± 2.9 SD	50.13 ± 1.50 SD	52.95 ± 2.13* SD	45.76 ± 2.58 SD	52.53 ± 2.55* SD

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(mgdL^{-1})					
Low-density lipoproteins (mgdL^{-1})	23.08 ± 1.84 SD	22.18 ± 2.24 SD	24.91 ± 1.53 SD	31.93 ± 1.96* SD	22.11 ± 1.70 SD	29.35 ± 1.66* SD

Note: All data represented as the mean ± SD (n=6). Values in the row with asterisk (*) are significantly different at p<0.05
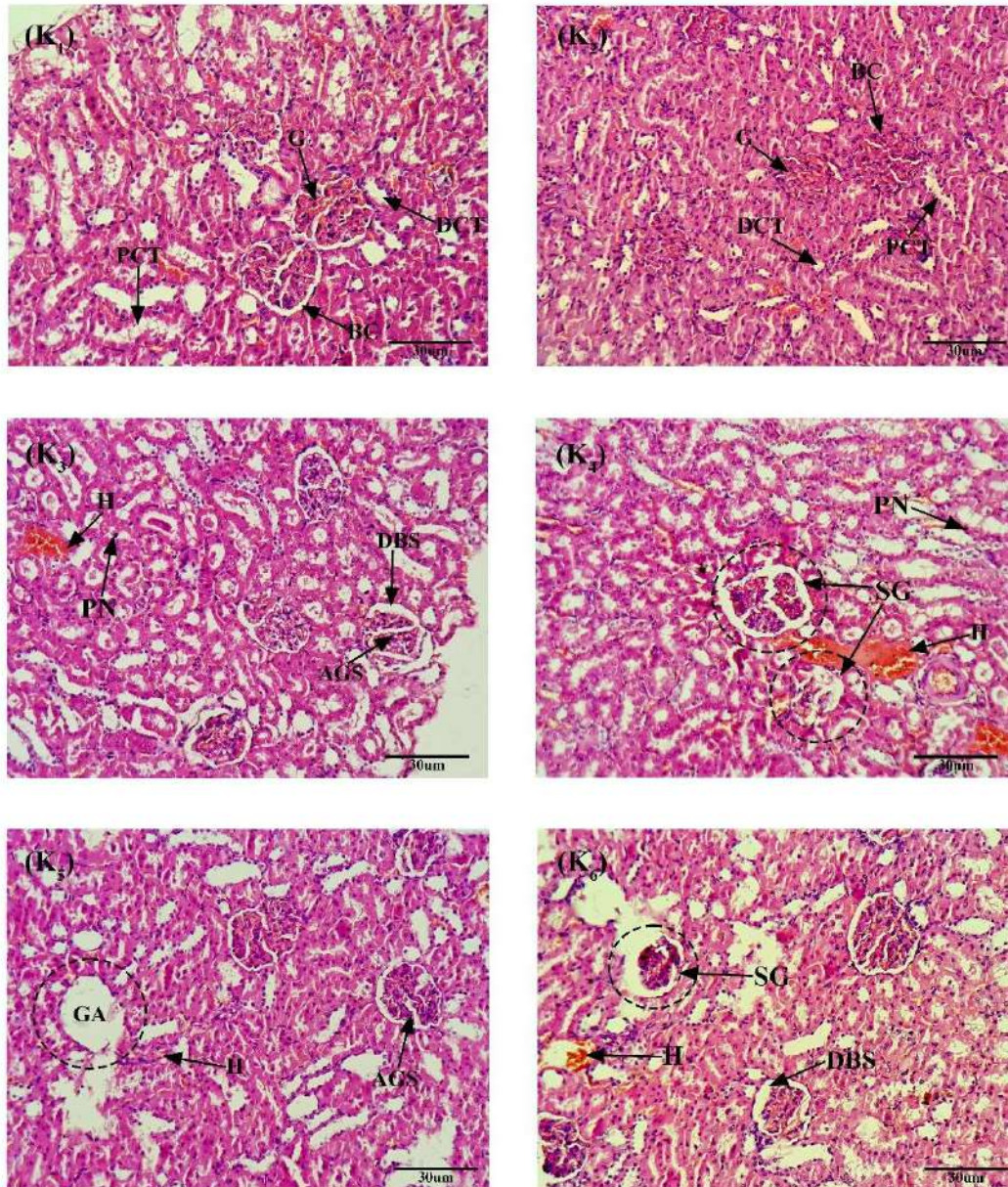
Fig. 1: Kidney sections of post experimental rat exposed to carbosulfan and *Cissus quadrangularis*.

RESEARCH ARTICLE

Quercetin Abrogates Graphene Oxide Induced Toxicity in Adult Zebrafish Model

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ABSTRACT

Due to remarkable properties such as a high specific surface area volume ratio and a low production cost, graphene oxides have lately emerged as a novel carbon-based nanoscale particle that offers an alternative path to graphene. While several studies have been conducted to evaluate the effects of GO on aquatic creatures, the ability of these organisms to recover following exposure to a flavonoid remains mostly unexplored. The present study examined the toxicity of graphene oxide (GO) in zebrafish (*Danio rerio*). The liver tissues were evaluated morphologically, and the optimal dose of quercetin for neutralizing unfavourable effects was determined. For fourteen days, fish were fed and five study groups were established: control, GO exposure alone (20 mg L^{-1}) and GO exposure mixed with three different quercetin doses (20 mg L^{-1} of GO+ quercetin 100, 150 and 200 mg kg^{-1} of food). *D. rerio* exposed to GO alone or in combination with a lower quercetin dose showed overall inferior results compared to control groups and those supplemented with GO and 150 or 200 mg kg^{-1} quercetin. The typical liver consisted of an uninterrupted mass of hepatocytes joined by an abundance of blood sinusoids. In the livers of fish exposed to GO, vacuolation, a loose organization of hepatic cells, histolysis and cell border disintegration were detected. As a result of our findings, we believe that sub-lethal doses of GO may be toxic to fish, harming the aquatic food chain. While GO is toxic to those exposed to it, quercetin supplementation at 150 or 200 mg kg^{-1}may act as a preventative strategy against its toxic effects.

Keywords: Graphene Oxide (GO), liver, oxidative stress, quercetin, zebrafish,

INTRODUCTION

Due to its remarkable properties, such as a high specific surface area volume ratio and a low manufacture cost, graphene oxide has lately emerged as a novel carbon-based nanoscale particle that provides an alternative route to graphene [1,2]. The graphene oxide is structurally identical to a graphene sheet, with its base including oxygen-containing groups such as hydroxyl, carbonyl, carboxylic and epoxy groups [3, 4]. Graphene is a two-dimensional material composed of hexagonally organized sp^2 hybridized carbon atoms, giving it a massive surface area on both sides of the planar axis [5]. The graphene family includes single-layer graphene, reduced graphene oxide, nanosheets, and graphene oxide (GO) [6- 9]. Graphene has been utilized in environmental, biomedical, and biotechnological applications [10-12]. While graphenes have made significant breakthroughs, if not revolutions, in the biomedical area, their use poses risks to human health, necessitating a greater level of nanotoxicological and human safety study. The extent to which graphene might cause toxicity in biological systems and the degree of safety connected with its use are critical issues to research [13,14]. Nanotechnology application in medicine has revolutionized drug formulation and delivery strategies. The notion of controlled-release systems has generated considerable interest because it enables an exact rate of release, enhanced bioavailability, and less unwanted effects in medicinal compounds [15-18]. Quercetin (3,5,7,3',4'-pentahydroxyflavone) is a flavonoid in various edible fruits, vegetable herbs, oils, and tea-related products, including apples, onions, and *Ginkgo biloba* red wine [19-21]. Even though research on quercetin's pharmacokinetics and bioavailability is sparse and variable, it has several benefits, including antioxidant, anticancer, and antibacterial characteristics, and as a result, it has garnered considerable interest [22].

Quercetin possesses various biological effects, including cardioprotection, anticancer, antiulcer, anti-allergic, anti-inflammatory, antiviral, and antiproliferative action [23-25]. Additionally, quercetin possesses antioxidant qualities and is beneficial in treating neurological disorders [26,27]. Quercetin is more powerful than the well-known antioxidants trollox, ascorbyl and rutin [28]. This occurs due to the quantity and location of free hydroxyl groups in the quercetin molecule [40]. As a result, quercetin's pharmacological properties, including anti-tumor and wound healing, have been extensively researched [29,30]. The zebrafish (*Danio rerio*), a well-known model animal, is used to conduct toxicity studies and to assess oral medicine delivery strategies [34]. Due to their genetic similarity to humans (about 70%), zebrafish have demonstrated physiological responses that are tremendously valuable for human health and therapy in toxicological and drug delivery investigations [31]. When injected intravenously into mice, GO caused granulomas in the lungs, liver, spleen, and kidney [32,33]. To better understand GO's potential toxicity, we examined its effect on oxidative stress responses in zebrafish. Understanding the effects of GO may aid in the safe deployment of this versatile material. Our findings aid in understanding the potential for GO to cause harm in aquatic environments.

MATERIALS

Graphene oxide synthesis using modified Hummer's method

1g graphite and 1g sodium nitrate were mixed in 46 mL sulphuric acid and stirred continuously for 2 hours in an ice bath. Additional 6g potassium permanganate, a strong oxidizing agent, was gradually added. The mixture was continuously stirred for two hours at 35°C until a brownish tint appeared. The suspension was then diluted with 100 ml deionized water stepwise fashion. Finally, 3 mL of hydrogen peroxide was added to the mixture, producing a yellow hue. To eliminate unreacted salt from the mixture, it was treated with 10% hydrochloric acid and centrifuged until neutral pH. Following centrifugation, a gel-like solution was generated subsequently dried for 12 hours at 60°C in a hot air oven to produce graphene oxide powder [36]. The synthesized GO was characterized via UV-spectrophotometer, FT-IR, X-ray diffractometer and SEM-EDX.

Fish and experimental conditions

We obtained 30 zebrafish (*Danio rerio*) weighing 1.93±0.2g from a local fish market in Kolathur, Chennai, Tamil Nadu. Before the trial, fish were acclimated for one week and fed commercial meals three times daily. The

photoperiod was maintained at 12L:12D and the water temperature was kept constant at 26.0±1.0°C. Daily measurements were done to analyze the physicochemical parameters: pH and dissolved oxygen levels. Five test groups were used in the experiment, namely: 0+0 - fish fed the control diet; 0+20 - fish fed the control diet+20 mg L^{-1} of GO;100+20 - fish fed the control diet+ quercetin (100 mg kg^{-1} of food) + 20 mg L^{-1} of GO; 150+20 - fish fed the control diet+ quercetin (150 mg kg^{-1} of food) +20 mg L^{-1} of GO; 200+20 - fish fed the control diet+ quercetin (200 mg kg^{-1} of food) +20 mg L^{-1} of GO.In 30 L aquariums, fish were randomly distributed. For 14 days, fish were fed twice a day, at 10:00 a.m. and 15:00. Sampling was done14 days following exposure.

Food preparation and Sampling

All groups consumed the same amount of food: 51.1 percent crude protein, 13.6 percent fats, 10.2 percent ash, and 3.55 percent fibre. A dose of GO (20 mg L^{-1}) was administered to the diet, as were three doses of quercetin (100, 150, or 200 mg kg^{-1} of food).

Histology

Histological examination of the liver was performed. After 24 hours of fixation in a 4% w/v paraformaldehyde solution, tissues were dried in ethanol, embedded in paraffin, sectioned and stained with hematoxylin and eosin. Each segment was examined under a light microscope.

Biochemistry

The zebrafish were rapidly euthanized in melted ice and the liver tissue was extracted as soon as possible. This was homogenized on ice with 200μL of ice-cold physiological saline before being centrifuged at 4000 g for 15 minutes at 4°C and the supernatant was collected. The quantities of glutathione (GSH) and malondialdehyde (MDA) in supernatants were measured using commercial kits and the manufacturer's procedure.

RESULTS

UV-Vis spectroscopy

The ultraviolet-visible absorption spectrum of GO is shown in Fig. 1. Maxima was seen between 200 and 800nm. GO found a high absorption peak around 216 nm, which corresponds to the oxygen-containing functional groups in GO, confirming the presence of GO in the carbon-based material produced.

X-Ray Diffraction (XRD)

Fig. 2 illustrates the XRD patterns of the synthesized carbon-based material. The diffraction exhibits a strong peak at an angle of 11.36°, 42.49°, which corresponds to the reflection planes a (002), (001), corroborating the synthesis of GO. The crystallite diameter (D) of the GO was calculated using the Debye–Scherrer equation (D = K/cos), where FWHM (full-width at half-maximum or half-width) is in radians, is the position of the diffraction peak's maximum, K is the so-called shape factor, which is typically around 0.9, and is the X-ray wavelength (1.5406 for Cu K). GO has a crystallite size of 31.5nm [35].

Fourier Transform Infra-Red (FT-IR) Spectroscopy

The FTIR spectrum was utilized to determine the functional groups present in the carbon material produced (Fig.3). Peaks were found between 400 and 4000cm^{-1}. The FT-IR peaks were observed at 2971.54cm^{-1} (O-H stretching), 1714.19cm^{-1} (C=O stretching vibration), 1386.59cm^{-1} (C-O deformation), 1224.73cm^{-1} (C-O stretching), 1032.01cm^{-1} (C-O stretching vibration), and 1582.76cm^{-1} (C=C stretching). The presence of functional groups containing oxygen indicates that the graphite has been successfully oxidized to GO.

SEM and EDX

The scanning electron microscope (SEM) is frequently used to examine the surface texture of graphene oxide (Fig.4). The GO appears crumpled and wrinkled under the SEM, with a hairy and gritty surface and a fuzzy sheet edge. The

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GO exhibited a soft carpet-like shape, resulting from the carboxyl and hydroxyl groups being attached to sheets. EDX tests (Table 1; Fig. 4(a)) revealed that graphene oxide is predominantly carbon and oxygen.

Histology

D. rerio exposed to GO alone or in combination with a lower quercetin dose showed inferior results than control groups and supplemented with GO and 150 or 200 mg kg^{-1} quercetin. In fig. 5, the typical liver consisted of an uninterrupted mass of hepatocytes joined by an abundance of blood sinusoids. In the livers of fish exposed to GO, vacuolation, a loose organization of hepatic cells, histolysis and cell border disintegration were detected.

Detection of oxidative stress indicators

As shown in Fig. 6 (a,b), exposure to GO altered MDA and GSH levels. MDA levels were significantly greater in fish exposed to 20 mg/L^{-1} GO than in control fish ($p<0.05$). As seen in Fig. 6c, GO exposure caused a significant ($p<0.05$) decrease in GSH levels, whereas the groups exposed to varied quercetin concentrations showed slow improvement in the levels of GSH. Supplementation of quercetin at different concentrations provided a protective role by maintaining the antioxidant level near normal to the control. (Fig. 6a and b). The groups exposed to varying concentrations of quercetin and GO showed significantly lower oxidative stress markers.

DISCUSSION

To use GO in real-world applications, extensive *in vitro* and *in vivo* studies demonstrating its biocompatibility and toxicity will be required. According to numerous research that used both *in vivo* and *in vitro* investigations, GO has toxicological implications [36,37]. Few studies have examined the effects of GO on aquatic species, owing to the complexity of natural aquatic systems in comparison to the smaller laboratory systems used in such toxicity studies. GO can aggregate or agglomerate in an aquatic environment depending on the pH, ionic strength, concentration, and dissolved organic matter [39,40]. Agglomeration or aggregation of GO can change its size, effective surface area, and other physicochemical features, influencing toxicity by making real-world exposure dosages more difficult to control. Higher GO concentrations were shown to be more prone to aggregation upon exposure. This may have influenced GO uptake, dispersion, and toxicity. These characteristics could explain why GO was hazardous to zebrafish independent of dosage in this study. Despite previous studies of the mechanisms driving GO toxicity, no conclusive data to support risk assessments or limitations were established [34]. Many nanoparticles are prone to toxicity due to oxidative stress. The production of reactive oxygen species (ROS) in target cells causes it. One mechanism of GO toxicity has been postulated to be the generation of reactive oxygen species (ROS) [22-25]. The balance between ROS generation and their elimination or reduction by antioxidant enzymes is reflected in normal cellular homeostasis, with GSH regulating ROS levels [40].

CONCLUSION

This study shows that GO caused oxidative damage in zebrafish at low concentrations of 20 mg L^{-1}. The liver underwent remarkable changes over time, demonstrating that the GO had a distinct effect on This tissue throughout the trial. On the other hand, it was shown that taking quercetin at a higher dose protected against GO exposure. Between 150 and 200mg kg -1 quercetin was the optimal dose for mitigating the adverse effects of GO exposure. The literature is deficient in information regarding how species recover from prolonged exposure. As a result of this finding, it is vital to learn more about how the body detoxifies itself following exposure to GO.

Conflict of interests

Authors declare no conflict of interests to disclose.

Ethical approval

There were no animal ethical issues involved in carrying out this research.

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Table.1 EDX measurement of graphene oxide

Elements	line	Mass%	Atom%
C	K	51.94±0.17	59.01±0.19
O	K	48.06±0.37	40.99±0.32
Total		100.00	100.00
GO		Fitting ratio 0.3127	
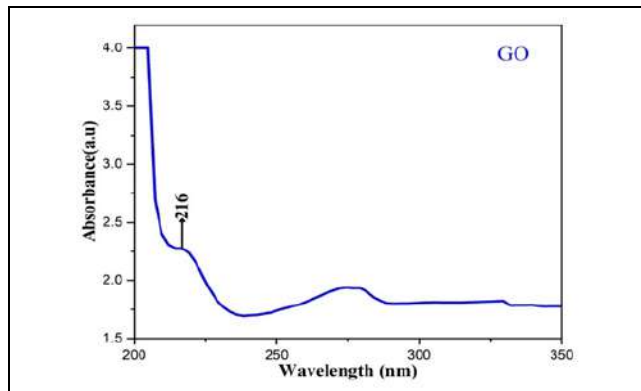
Fig.1 UV-vis absorption spectra of GO
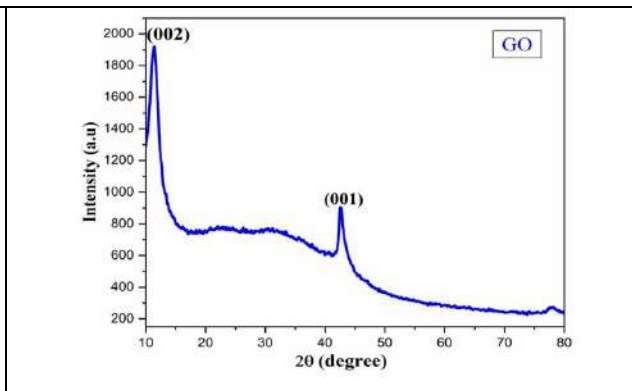
Fig.2 XRD pattern of GO
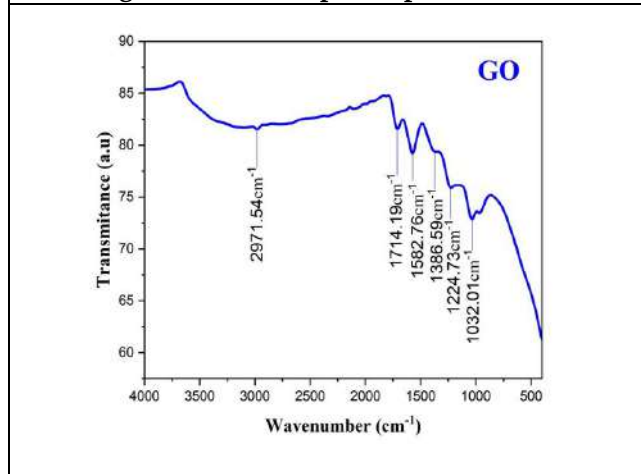
Fig. 3 FTIR spectra for GO
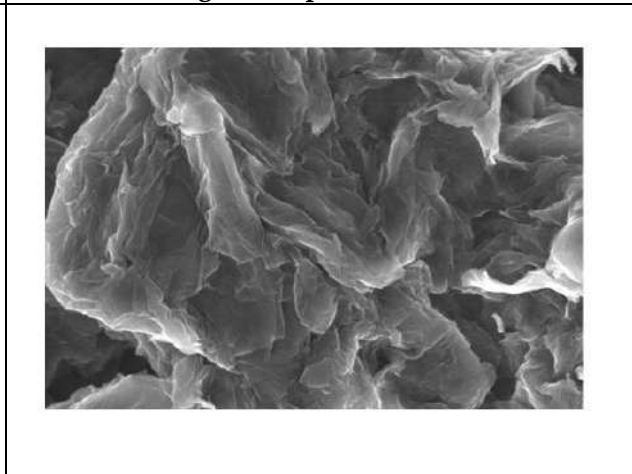
Fig.4 Scanning Electron Microscopy image for GO
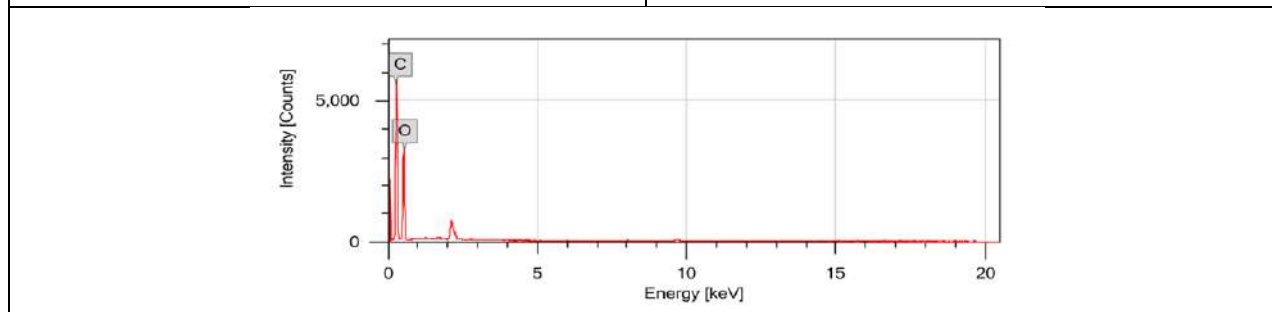
Fig. 4(a) Energy Dispersive X-ray analysis for GO

Ragunath Ravichandiran *et al.,*
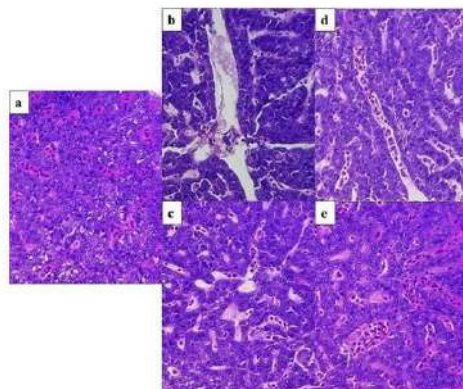
Fig.5 Liver morphology in adult zebrafish (*Danio rerio*) following exposure to graphene oxide nanoparticles for 14 days. (a) control group exhibits normal histology; (b) treatment with 20 mg L⁻¹ resulted in peripherally located nuclei, pyknotic nuclei and vacuole formation (VF); (c,d, and e) treatment with quercetin resulted in the decreased number of swollen cells (SC); and necrosis (N).
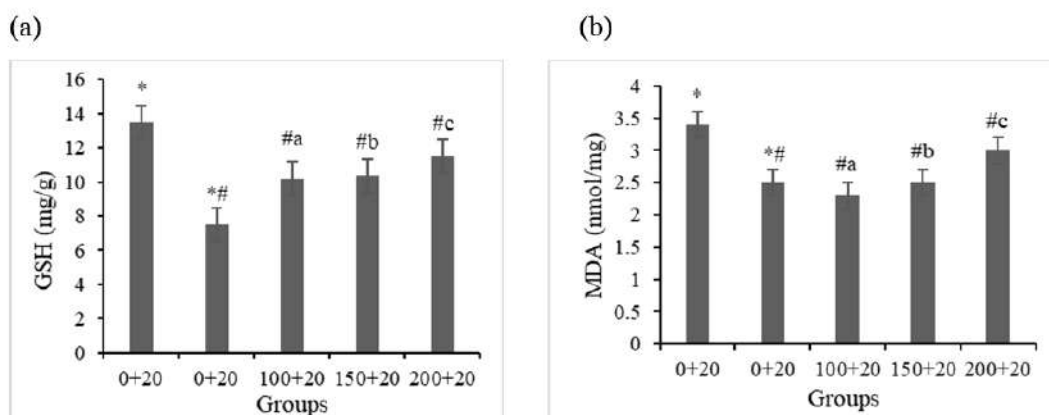
Fig.6 Oxidative stress biomarkers in the liver of *Danio rerio* a) GSH activity (mg/g); b)MDA (nmol/mg); 0+0, 0+20, 100+20, 150+20, 200+20 indicate the test groups. Data are expressed as mean± standard error of means. Different letters indicate the significant differences between the groups ($p<$ 0.05).

RESEARCH ARTICLE

Effect of Trunk Balance on Motor Recovery, Gait Velocity and Functional Balance in Recovering Stroke Patients - A Correlation Study

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ABSTRACT

Stroke or brain attack is sudden loss of neurological function caused by sudden interruption of blood supply to brain result in neurological deficit. As a result of stroke there will be loss of strength in trunk and leg muscles as well as balance that may disturb walking ability. Gait parameters specifically gait velocity is also affected as a consequences of stroke. Affection of trunk balance will affect motor recovery and functional Balance. The study is conducted to find out relationship between the trunk balance with motor recovery, gait velocity and functional balance in recovering stroke patients. Total 32 patients were screened as per inclusion and exclusion criteria. Demographic data were taken from each patient.At the time of discharge from the hospital trunk balance was taken with trunk impairment scale. Motor recovery was assessed with use of Fugl Mayer scale. 6 min walk test was taken to measure gait velocity and functional balance was taken with use of Berg Balance scale. Total 32 Recovering stroke Patients have participated in the study.All the stroke patients who have participated in the study were having mean age of 52.6 years and participants of both the genders were included .Amongst the participants' males were 78.21% and females were 21.9%. Participants with both sided strokes were included in the study. The result found Weak negative correlation between TIS score with Gait Velocity and Gait Velocity with Berg Balance scale. Weak positive correlation were establish between TIS score and Fugl-Mayer Score and TIS with Berg Balance scale. The result found weak positive correlation between Gait velocity and Fugl-Mayer Score and Fugl-Mayer Score with Berg Balance scale score. Trunk Balance haslittle impact on motor recovery and functional balance but will not have much influence on gait velocity. Motor recovery influence gait velocity and functional balance.

Keywords: Functional Balance, Gait velocity, Motor Recovery, Stroke, Trunk Balance.

INTRODUCTION

Stroke or brain attack is sudden loss of neurological function caused by sudden interruption of blood supply to brain; ischemic stroke occurs due to clot which blocks the vessel or as a result of impairment in blood flow which produce deprivation in oxygen and nutrients supply to brain. Haemorrhagic stroke takes place due to rupture of blood vessel causing leakage of blood in the brain [1] Cerebrao Vascular Stroke has been found as one of the major cause of death and disability in all community dwelling individuals [2]. Stroke survivors commonly have impaired motor and postural control mechanism due to lack of interaction between sensorimotor and cognitive functions which will result in impairment of trunk control [3]. Stroke is very important causes of death and debility in India [4]. Motor difficulties are one of the most common and disabling of stroke patients' shared incapability. Hemiparesis is a prevalent condition that requires immediate treatment. The degree of the hemiparesis affects the patient's functional abilities as well.

Independence In stroke patients, walking plays a critical role in determining functional recovery. Gait is a critical indicator of self-sufficiency. As a result, it's no surprise that improving walking capability is the most often mentioned priority for stroke survivors. Balance issues are the most prevalent neurological weakness after a stroke, and they cause poor recovery of activities of daily living (ADL), movement, and may also increase the risk of falling in stroke survivors [5]. "Inability to maintain an upright position within the limitations of stability or base of support," according to the definition of balance disability. Studies of balance impairments demonstrate that there is an altered weight distribution pattern in which the weak leg bears less weight and has a lower risk of falling. The pattern mentioned above can be seen in all elements of balance, including static, dynamic, and responses to external perturbations, and it's more frequent in stroke survivors with high functional levels, such as community ambulatory. Some research have shown that measurements of balance impairment are related to activity level, whereas others have failed to show this relationship.

The above-mentioned findings could be due to a variety of factors. Balance issues have been linked to a slower recovery of functions such as activities of daily living (ADL) and mobility, which increases the risk of falling [6]. They have reduced ROM while shifting their weight around the fix base of support, especially in the direction of the weaker leg, due to a balance problem that causes uneven weight distribution on the lower limb. As a result, less weight bearing occurs on the weak leg, and they have reduced ROM while shifting their weight around the fix base of support [7]. Stroke will result in paresis or paralysis of one side of body which produce reduce ability of trunk for adjustment and decrease trunk muscle activation while performing voluntary movement and reduce area of stability which affect Equilibrium reaction. In addition to these trunk muscle spasticity specifically in muscles of shoulder and the pelvic girdle produce pelvic inclination in upward direction and scapular downward rotation, which produce trunk lateral flexion [11]. Decrease weight bearing on affected limbs will increase risk of fall [12].

Walking dysfunction, which is followed by stroke1, is the most usually mentioned functional constraint, and it will impair objectivity, quality of life, and participation in various activities. According to popular belief, gait improves during the first 3 to 6 months after a stroke and then stabilises. Additionally, continue with limits for walking after a stroke, such as increased energy expenditure, balance control, and musculoskeletal injury of the non-paretic lower extremity, which may limit their activity despite their ability to walk independently. Wade and colleagues explain a reduction in mobility following a stroke as a result of this. Gait is a significant aspect in achieving self-sufficiency. That is why improving walking function is a top objective for stroke patients who have residual deficits. After a stroke, there are several ways to assess balance. Due to impaired weight bearing on the affected lower leg and stiffness, people who have had a stroke are more likely to fall. Falls can both be predicted and avoided. Falls are common if sufficient safety precautions are not performed. Falls are common during hospital stays and can have negative consequences that require specific attention. As a result, it's critical to determine which stroke survivors are at risk of falling and to provide fall prevention strategies. This can be accomplished by doing a thorough examination of the patients using the most regularly used clinical tests that are both valid and reliable. Many

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countries have used the use of statistical models to predict the danger of a fall like Time up and Go test, Berg Balance scale and 6 minute walk test.TIS is used to measure the trunk balance and Impairment after stroke. . The TIS assesses both static and dynamic balance in sitting position. It also evaluates trunk co-ordination while performing movement. The TIS has satisfactory reliability, validity and internal consistency for clinical practice and is widely use in research related to stroke [8]. Gait velocity is one of the two most important gait specific parameters applicable to the stroke population and is a mostly used measure for the gait performance and can be used to discriminate the levels of disability in the stroke survivors. The 6-minute gait test was used primarily as a simple gait test to determine ability and endurance in activities of daily living in Stroke survivors. In one of the study performed recently, it has been suggested that distance travelled at 6MWT can be used as a clinical measure to assess walking ability in patients who have had a stroke. The reliability of the 6MGT test and retest in stroke patients was found to be good. Therefore, the distance travelled during the 6MWD is evaluated to be clinically sensitive to counter the changes in stroke survivors.

The Fugl-Meyer scale quantitative evaluation tool for assessing sensorimotor stroke recovery, based on fundamentals of Brunnstrom stages of motor recovery in stroke survivors with Hemiplegia. It comprises 100-point for motor domain. It is having good construct validity reliability (interrater and intrarater) and have been demonstrated, and preliminary evidence suggests that the Fugl-Meyer assessment is responsive to change [9]. The BBS is a 14-item scale that quantitatively assesses balance and risk for falls in older community-dwelling adults through direct observation of their performance. A global score is calculated out of 56 possible points. Scores of 0 to 20 represent balance impairment, 21 to 40 represent acceptable balance, and 41 to 56 represent good balance. The BBS measures both static and dynamic aspects of balance [10].

Need of the Study
Trunk balance is often found affected in stroke patients but in the rehabilitation process it is often neglected which may affect ultimate outcome related to stroke recovery. So, if any association found between trunk balance and motor recovery, gait velocity and functional balance then during rehabilitation process emphasis will be provided on trunk balance which may help in motor recovery, functional balance and improvement in Gait velocity.

METHODOLOGY

The study was approved by Ethical committee of Parul University (PU IECHR).Data was collected from the stroke patients referred for Physiotherapy from IPD and Physiotherapy OPD of Parul Sevashram Hospital. Testing sessions were performed at Neurophysiotherapy department of Parul Sevashram Hospital. Stroke Patients included in the study were assessed for cognitive functions and the Walking capability. Both Male and female patients between 40 to 70 years with ischemic or haemorrhagic stroke for the first time with 6 month or more than that duration were included in the study. Participant demographics and stroke characteristics such as age, gender, side of lesion, side of Hemiparesis and type of stroke (ischemic or haemorrhagic), Hand dominance were recorded at the time of interview. Mini Mental state Examination (MMSE) was use to assess cognitive level of the stroke survivors. Patients were also assessed for Brunnstrom stages of recovery and Patients with Brunnstrom stage 3or more than that were included in the study. Patient who have shoulder problem or other Psychiatric issues, severe cardiorespiratory problem or epilepsy were not included in the study. 6MWT was taken as a screening tool for gait ensuring participants could walk safely and independently. Steps taken in the stipulated time was noted with a stopwatch; steps taken and gait velocity was calculated by the formula distance/time.

Data Management and Analyses
Demographic, Score of FuglMayer and BBS scale and values of gait Velocity were entered into a Microsoft Excel prior to analysis. The individual gait parameters from each trial on the walkway were recorded. Statistical analysis was performed in SPSS (version 20).

Descriptive analyses were used to present demographic characteristics. Multiple correlation analyses were performed to examine the effect of Gait Parameters. Pearson correlations were used to check for correlations between gait velocity and Stride Length. Pearson correlations were used to check for correlations between Gait Velocity and Stride Length with Balance and Quality of Life

RESULT

Correlation was calculated on Total 32 Recovering stroke Patients who were recruited for study. Participants had a mean age of   52.65 years, and included both males and females (78% male and 22% female). Individuals presented with both left (18.70%) and right-sided (81.25%) strokes. The result found Weak negative correlation between TIS score with Gait Velocity(r= -0.2682) and Gait Velocity with Berg Balance scale (r=-0.275). Weak positive correlation were establish between TIS score and Fugl-Mayer Score(r= 0.1237.) and TIS with Berg Balance scale (r=0.2024). The result found very weak positive correlation between Gait velocity and Fugl-Mayer (r=0.0802.)Score and Fugl-Mayer Score with Berg Balance scale score (r= 0.2935.).

DISCUSSION

In this study data was analysed for 32 patients to find the relationship of Trunk balance with Motor recovery, Gait velocity and Functional Balance. Jijimol G and Fayaz RK [13] discovered that trunk performance and static or dynamic balance are associated, and that any measure to increase trunk performance will improve balance in stroke patients. Trunk stability is thought to be an important component of balance and coordinated extremity usage in daily functional activities and higher-level motor tasks. Due to this reason positive relation has been establish between TIS with BBS and Fugl-Meyer Score. Exercises for the trunk rehabilitation of stroke patientscan be use to enhanced balancing activities needed for standing and walking, hence enhancing efficient motor strategies and will produce improve motor recovery. Experts in the field of neurological rehabilitation have identified the trunk as the body's core important point, with movement control progressing from the proximal to distal body parts. According to the literature on motor control, if proximal trunk control gains are achieved, enhanced distal limb control can be expected during balancing and functional mobility [14]. Due to this reason positive relation has been found between TIS and BBS.

Goldie et al [15] reported that the improvement in trunk control that comes with trunk rehabilitation may be the cause of the shift in gait speed. As a result, if proximal trunk control is increased, distal lower extremity mobility, such as that required for walking, should be enhanced as well. Other study found that trunk control is not important component in deciding functional outcomes if patient has intact gait ability after stroke. According to this study mobility will improve but gait speed may or may not improve due to that weak negative correlation is established in this study. In one of the study it was found that "the total score of TIS was the only predictor of the motor function of the lower limbs and the dynamic balance of TIS was a predictor of the motor function of the upper limbs." The relative contributions of standing balance and trunk balance to gait ability and motor.Function were revealed in this study. They show that balance has a high power of explanation for gait ability and that trunk balance, rather tha n gait ability, is a determinant of motor Function. Improvement in trunk muscle activation will produce improve postural control, and gait speed in hemi paretic stroke patients. As per one of the study "The clinical balance scores are analogous to walking speed, which is a global indicator of gait dysfunction, but it does not identify potential contributing factors" M. A. Dettmann et al.[i] in his study assessed gait parameters and stability in a weight bearing position and found that hemi paretic patients recompense for a loss of balance with smaller step length and reduced Gait speed. Which will reduce Gait velocity but maintain Balance.

CONCLUSION

In the present study, it is found that Trunk Balance haslittle impact on motor recovery and functional balance but will not have much influence on gait velocity. Motor recovery influence gait velocity and functional balance. Hence, improvement in Trunk balance should be consider to improve functional balance and promote motor recovery.

Limitation:
1. All the variables affecting Gait and balance are not considered in the study.
2. Overall motor recovery was considered as total score of Fugl-Meyer score.
3. Division of patients according to age group was not done in the study.

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Table-1 Participant's Characteristics

Participant's Characteristics:	Mean	SD
Age	52.65	3.87
Gender		
Male	78%	0.40
Female	22%	
Side of stroke		
Left	18.70%	0.44
Right	81.25%	
MMSE score	28.12	0.90
Gait velocity	.28	0.04
Trunk Impairment Score	11.68	1.30
Berg Balance scale Score	38.34	4.44
Fugl Mayer Score	81.65	7.90

Table-2 Correlation between Trunk impairment scale, Berg Balance scale, Gait velocity and Fugl Mayer scale

		TIS	BBS	Gait velocity	Fugl Mayer Score
TIS	Pearson Correlation	1	0.2024	-0.2682.	0.1237.
	Sig. (2-tailed)	.			
	N	32	32	32	32
BBS	Pearson Correlation	-.221	1	-0.275.	0.2935.
	Sig. (2-tailed)	.225	.		
	N	32	32	32	32
Gait velocity	Pearson Correlation	-0.2682.	-0.275.	1	0.0802.
	Sig. (2-tailed)			.	
	N	32	32	32	32
Fugl Mayer Score	Pearson Correlation	0.1237.	0.2935.	0.0802.	1
	Sig. (2-tailed)				.
	N	32	32		32

Figure-1 Gender Distribution and Side of Lesion	**Figure-2 TIS and Gait Velocity** **The Graph shows weak negative correlation between TISscore and Gait Velocity.**

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Figure-3 TIS and Fugl-Mayer Score
The Graph shows weak positive correlation between TISscore and Fugl-Mayer Score.

Figure-4 TIS and Berg Balance scale
The Graph Shows weak positive correlation betweenTISscore and Berg Balance scale

Figure-5 Gait velocity and Fugl-Mayer Score
The Graph Shows weak positive correlation between Gait velocity and Fugl-Mayer Score.

Figure-6 Gait Velocity and Berg Balance scale
The Graph shows weak negative correlation between Gait Velocity and Berg Balance scale score.

Figure-7 Fugl-Mayer Score and Berg Balance scale
The Graph Shows weak positive correlation between Fugl-Mayer Score and Berg Balance scale score.

RESEARCH ARTICLE

Validation of Hot Air Oven using RTD & Thermocouple Sensors

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ABSTRACT

The aim of the present study was to perform validation of a Horizontal Dry heat sterilizer located in a Pharmaceutical Quality Assurance PG laboratory at JSS College of Pharmacy, Mysuru. The study was mainly focused on operational and performance qualification. Dry heat is used to assure that it is free from pyrogenic material for glass and other laboratory equipment. The sterilization inside a chamber is a vital process and there is a mandatory regulation for validation process globally. For a fixed period of time, the sterilizer is used to heat all parts of its load to the specific temperature, long enough to achieve the desired sterility. Empty chamber heat distribution was carried out to check for absence of cold spots and to check for uniform heat distribution, in order to provide a sterilizing environment. Hence the maximum & minimum value of average for heat distribution study was found to be 153.7ᵒC & 150.76ᵒC (RTD), 150.47ᵒC & 146.82ᵒC (Thermocouple) respectively. During the performance qualification, loaded chamber heat penetration test was carried out on various loads, hence the maximum & minimum value of average for heat penetration study was found to be 150.89 & 150.33ᵒC (RTD), 150.40ᵒC & 147.55ᵒC (Thermocouple) respectively. Therefore, it can be concluded that, the Dry heat sterilizer was validated and ready for the use.

Keywords: Validation, Dry heat sterilizer, operational qualification, performance qualification.

INTRODUCTION

Validation: Establishing documented evidence that provides a high degree of assurance that a specific equipment, services, materials and process will consistently produce a product meeting its predetermined specification and quality characteristics [1].

Hot Air Oven: Hot air oven is an electrical device which is used for dry heat sterilization. The equipment exposed to dry heat sterilization must be dry and in case any product is used it shouldn't melt at higher temperature. Usually powders with high melting point is sterilized using dry heat sterilization method [2].

Temperature Mapping: Temperature mapping is the process of validating the ability of a storage area to maintain the temperature in the specified limit. Temperature mapping is usually considered for fridge, freezers, cold rooms, storage areas, autoclaves and hot air ovens, etc. in the process of temperature mapping sensors are distributed throughout the instrument or the room in the pre-defined locations. The temperature on the external sensors should be within the specified limit for the instrument or the room to qualify the temperature mapping [3].

Temperature Sensors

Thermocouples: A thermocouple uses two metal wires to produce a voltage relative to the temperature present in the junction between them. There are many specialized kinds of thermocouples – they can combine different metals to measure various characteristics and temperature ranges, and produce specialized calibrations.

Resistance temperature detectors (RTDs): An RTD sensor measures temperature based on the resistance changes in a metal resistor inside. The most popular RTDs, called PT100 sensors, use platinum and have a resistance of 100 ohms at 0°C [4].

MATERIALS AND METHODS

List of Materials used
➢ Calibrated data logger
➢ Calibrated sensors
▪ RTD Sensors
▪ Thermocouple Sensors
➢ Hot air oven
➢ Dummy load

Methods
Empty Chamber Heat Distribution Study
➢ Empty chamber cycle is carried out to study heat distribution profile in the hot air oven chamber.
➢ Place minimum of 6 calibrated temperature sensors in the identified worst-case location based on the logical assessment of the chamber design. Diagram of placement of temperature sensor for empty heat distribution study as per Table No. 4
➢ The probe tips should be suspended to avoid contacting any solid surface. Keep chemical indicator near the probe.
➢ Operate the dry heat sterilizer as per the SOP and start the data logger to record the sensor temperature within the chamber with respect to time. The temperature data shall be captured every 10 second interval.

The following details about the cycle shall be mentioned in the report.
• Mapping start time
• Mapping end time
• Minimum temperature during the study
• Maximum temperature during the study
• Minimum average temperature
• Maximum average temperature
Record the temperature data and the dry heat sterilizer operating parameters in the report.

Loaded Chamber Heat Distribution Study

➢ Temperature distribution profile with the loaded chamber shall be performed to determine the cold spot when the chamber is loaded.
➢ The sensors are placed as shown in the empty chamber heat distribution study
➢ While placing the sensor make sure that the sensor tips does not touch any surface or walls of the chamber.
➢ Description of the item loaded in particular load is mentioned in table no.1
➢ Take a photograph of load configuration and attach it with validation report
➢ Operate the dry heat sterilizer as per the SOP and start the data logger to record the sensor temperature within the chamber with respect to time.
➢ The temperature data shall be captured every 10 second interval.
➢ The following details about the cycle shall be mentioned in the report:
  • Mapping start time
  • Mapping end time
  • Load number
  • Minimum temperature
  • Maximum temperature
  • Maximum average temperature
  • Minimum average temperature

Door Open Recovery study

➢ Door open recovery study is performed to find the
➢ Sensors are placed as shown in the empty chamber heat distribution study.
➢ While placing the sensor make sure that the sensor tips does not touch any surface or walls of the chamber.
➢ Operate the dry heat sterilizer as per the SOP and start the data logger to record the sensor temperature within the chamber with respect to time.
➢ The temperature data shall be captured every 10 second interval.
➢ Wait for the dry heat sterilizer to stabilize. The time taken for stabilization of temperature can be understood from the previous studies.
➢ Once the hot air oven is stabilized note the time and open the door for about 2 minutes. (2 minutes are considered as a worst-case scenario).
➢ After completion of 10 minutes, note the time again and wait for about 30 minutes to 1 hour to ensure all the sensors achieved chamber condition.

The following details about the cycle shall be mentioned in the report:
• Mapping start time
• Mapping end time
• Minimum temperature
• Maximum temperature
• Maximum average temperature
• Minimum average temperature
Study the temperature profile and conclude the maximum time allowed for door open conditions.

Power failure recovery study

➢ The power failure recovery study is carried out to determine the maximum time that the instrument can operate in power failure condition.
➢ Sensors are placed as shown in the empty chamber heat distribution study.
➢ While placing the sensor make sure that the sensor tips does not touch any surface or walls of the chamber.
➢ Operate the dry heat sterilizer as per the SOP and start the data logger to record the sensor temperature within the chamber with respect to time. The temperature data shall be captured every 10 second interval.

➢ Allow the hot air oven to stabilize.
➢ After stabilization record the time and switch off the power supply to the equipment
➢ After 10 minutes switch on the power supply to the equipment and note the time taken for the instrument to stabilize.

The following details about the cycle shall be mentioned in the report
- Mapping start time
- Mapping end time
- Minimum temperature
- Maximum temperature
- Maximum average temperature
- Minimum average temperature

RESULTS

Stabilization of Hot air oven
Starting time: 12: 28 PM
Starting Temperature: 38°C
End Time: 12: 45 PM
End temperature: 151°C
Stabilization Time: 1 Hour and 20 Minutes approximately.

SUMMARY
Based on the data evaluation of the study, following were the interpretations.

1 Hour Empty chamber heat distribution Study (RTD sensors)
➢ Minimum temperature was observed to be 149.6 °C at sensor no. RTD 1.
➢ Maximum temperature was observed to be 154.2 °C at sensor no. RTD 5.
➢ Minimum average temperature was observed to be 150.76 °C at sensor no. RTD 1.
➢ Maximum average temperature was observed to be 153.7 °C at sensor no. RTD 5.
➢ Time taken for stabilization was found to be 1 hour and 20 minutes

1 Hour Empty chamber heat distribution study (Thermocouple)
➢ Minimum temperature was observed to be 145.4 °C at sensor no. Thermo 6.
➢ Maximum temperature was observed to be 151.8 °C at sensor no. Thermo 1.
➢ Minimum average temperature was observed to be 146.82 °C at sensor no. Thermo 5.
➢ Maximum average temperature was observed to be 150.47 °C at sensor no. Thermo 1.
➢ Time taken for stabilization was found to be 1 hour and 20 minutes

2 hours loaded chamber heat distribution study (RTD)
➢ Minimum temperature was observed to be 147.2 °C at sensor no. RTD 6.
➢ Maximum temperature was observed to be 152.7 °C at sensor no. RTD 1.
➢ Minimum average temperature was observed to be 150.33 °C at sensor no. RTD 2.
➢ Maximum average temperature was observed to be 150.89 °C at sensor no. RTD 4.
➢ Time taken for stabilization was found to be 1 hour and 20 minutes

2 hours loaded chamber heat distribution study (Thermocouple)
➢ Minimum temperature was observed to be 147 °C at sensor no. Thermo 2, 3, 4, 5, 6.
➢ Maximum temperature was observed to be 152.7 °C at sensor no. Thermo 1.
➢ Minimum average temperature was observed to be 147.55 °C at sensor no. Thermo 5.

➤ Maximum average temperature was observed to be 150.40 °C at sensor no. Thermo 1.
➤ Time taken for stabilization was found to be 1 hour and 20 minutes

90 seconds Door open and recovery study (RTD)
➤ Minimum temperature was observed to be 122.1 °C at sensor no. RTD 5.
➤ Maximum temperature was observed to be 153.5 °C at sensor no. RTD 5.
➤ Minimum average temperature was observed to be 142.36 °C at sensor no. RTD 5.
➤ Maximum average temperature was observed to be 147.79 °C at sensor no. RTD 4.
➤ Time taken for recovery was found to be 14 minutes and 45 seconds.

90 seconds Door open and recovery study (Thermocouple)
➤ Minimum temperature was observed to be 138.3 °C at sensor no. Thermo 4.
➤ Maximum temperature was observed to be 153.3 °C at sensor no. Thermo 1.
➤ Minimum average temperature was observed to be 145.67 °C at sensor no. Thermo 5.
➤ Maximum average temperature was observed to be 149.76 °C at sensor no. Thermo 1.
➤ Time taken for recovery was found to be 7 minutes and 20 seconds.

10 minutes power failure recovery study (RTD):
➤ Minimum temperature was observed to be 144.7 °C at sensor no. RTD 1.
➤ Maximum temperature was observed to be 153.9 °C at sensor no. RTD 5.
➤ Minimum average temperature was observed to be 148.26 °C at sensor no. RTD 1.
➤ Maximum average temperature was observed to be 151.06 °C at sensor no. RTD 5.
➤ Time taken for recovery was found to be 17 minutes

10 minutes power failure recovery study (Thermocouple)
➤ Minimum temperature was observed to be 14°C at sensor no. Thermo 6.
➤ Maximum temperature was observed to be 152.3°C at sensor no. Thermo 1.
➤ Minimum average temperature was observed to be 144.91 °C at sensor no. Thermo 5.
➤ Maximum average temperature was observed to be 148.28 °C at sensor no. Thermo 1.
➤ Time taken for recovery was found to be 6 minutes and 40 seconds

CONCLUSION

The validation of the hot air oven was carried out successfully with heat distribution study including empty chamber heat distribution and loaded chamber heat distribution study along with worst case study including 90 seconds door open and recovery study and 10 minutes power failure and recovery study. The results were found to be good and well within the acceptance criteria.

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Table 1: Load pattern for loaded chamber heat distribution study

Load	Dimension	Number
Conical flask	250ml	16
Glass bottles	500ml	20
Spatulas	--	--

Table 2: Details of Loggers and Sensors used

Sl. No.	Unit	Make	Model	Serial No.	Calibration done on	Calibration due on	Certificate traceability
1.	Datalogger	Brain child	PR 20	186110012	06/08/2021	05/08/2022	1701/L/655- 01
2.	Sensors (RTD Pt-100)	-	-	-	06/08/2021	05/08/2022	1701/L/656-02
3.	Sensors (Thermocouples K Type)	-	-	-	06/08/2021	05/08/2022	1701/L/658-04

Table 3: List of Sensor Locations

Sl. No.	Sensor No.	Location of sensors in hot air oven	Location description
1.	**Sensor 1**	1	Front Left Corner (Rack 1)
2.	**Sensor 2**	2	Back Right Corner (Rack 1)
3.	**Sensor 3**	3	In Front of Inbuilt Sensor
4.	**Sensor 4**	4	Back Left Corner (Rack 3)
5.	**Sensor 5**	5	Front Right Corner (Rack 3)
6.	**Sensor 6**	6	In Centre (Rack 2)

Table 4: Empty chamber heat distribution study (RTD)

Sensor No.	Temperature in °C		
	Minimum	**Maximum**	**Average**
RTD 1	149.6	151.6	150.76
RTD 2	151.6	153.3	152.74
RTD 3	150.1	151.9	151.23
RTD 4	150.1	152.1	151.36
RTD 5	152.9	154.2	153.70
RTD 6	150.5	152.3	151.72

Table 5: Empty chamber heat distribution study (Thermocouple)

Sensor No.	Temperature in °C		
	Minimum	**Maximum**	**Average**
Thermocouple 1	149.1	151.8	150.472
Thermocouple 2	148.3	150.7	149.633
Thermocouple 3	147.1	149.8	148.519
Thermocouple 4	146.4	148.8	147.688
Thermocouple 5	145.5	148.1	146.827
Thermocouple 6	145.4	149	147.067

Table 6: Loaded chamber heat distribution study (RTD)

Sensor No.	Temperature in °C		
	Minimum	**Maximum**	**Average**
RTD 1	148.1	152.7	150.5567
RTD 2	148.4	151.9	150.335
RTD 3	148.3	151.9	150.4086
RTD 4	147.8	151.9	150.89
RTD 5	150	151.9	150.5735
RTD 6	147.2	151.9	150.7037

Table 7: Loaded chamber heat distribution study (Thermocouple)

Sensor No.	Temperature in °C		
	Minimum	**Maximum**	**Average**
Thermocouple 1	147.5	152.7	150.4087
Thermocouple 2	147	151.5	149.5146
Thermocouple 3	147	150.6	148.4958
Thermocouple 4	147	149.7	147.9099
Thermocouple 5	147	148.9	147.5559
Thermocouple 6	147	149.6	147.7049

Table 8: Door open and recovery study (RTD)

Sensor No.	Temperature in °C		
	Minimum	**Maximum**	**Average**
RTD 1	141.3	150.8	147.5929
RTD 2	138.3	152.7	147.3054
RTD 3	135.2	151.3	145.6743
RTD 4	140.2	151.4	147.7963
RTD 5	122.1	153.5	142.3631
RTD 6	136.7	151.7	146.1212

Table 9: Door open and recovery study (Thermocouple)

Sensor No.	Temperature in °C		
	Minimum	**Maximum**	**Average**
Thermocouple 1	143.8	153.3	149.7635
Thermocouple 2	143	152	148.7519
Thermocouple 3	142	151.5	147.9442
Thermocouple 4	138.3	149.9	145.9
Thermocouple 5	139.6	149.4	145.6712
Thermocouple 6	141.9	150.6	146.7538

Table 10: Power failure recovery study (RTD)

Sensor No.	Temperature in °C		
	Minimum	**Maximum**	**Average**
RTD 1	144.7	150.8	148.2643
RTD 2	147.8	153	150.4758
RTD 3	146.3	151.6	149.0571
RTD 4	146	151.5	149.1207
RTD 5	148.2	153.9	151.0697
RTD 6	147	152.1	149.5378

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Table 11: Power failure recovery study (Thermocouple)

Sensor No.	Temperature in °C		
	Minimum	**Maximum**	**Average**
Thermocouple 1	144.2	152.3	148.2812
Thermocouple 2	143.9	151.3	147.583
Thermocouple 3	142.8	150	146.5382
Thermocouple 4	142.2	149.1	145.763
Thermocouple 5	141.3	148.2	144.9194
Thermocouple 6	141	148.9	145.0491
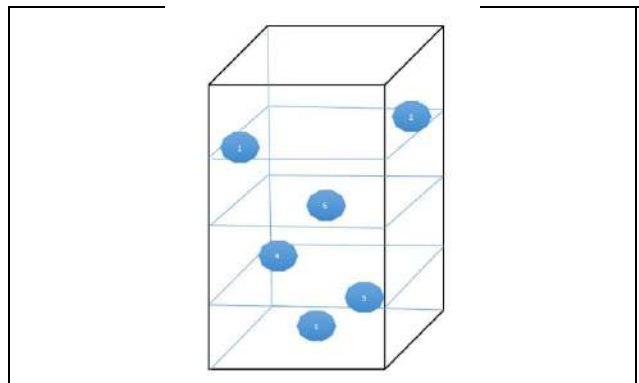
Figure 1: Schematic representation of sensors location inside the hot air oven
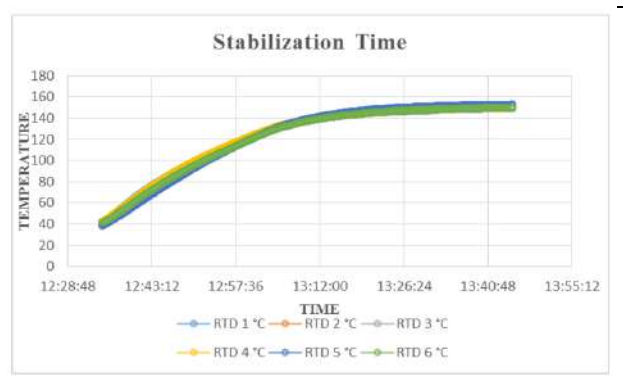
Figure 2: Stabilization of Hot air oven
1 Hour Empty chamber heat distribution Study (RTD sensors)
Starting time: 12: 50 PM
End time: 01: 50 PM
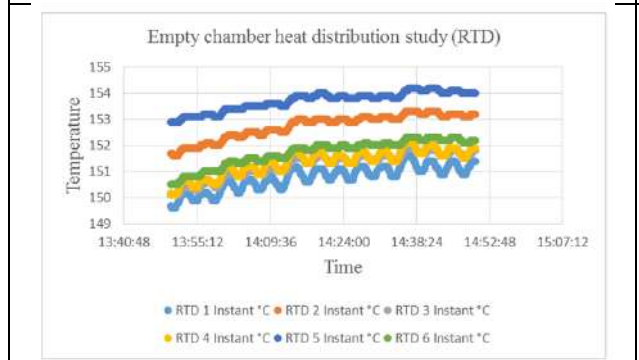
Figure 3: Empty chamber heat distribution study (RTD)
1 Hour Empty chamber heat distribution study (Thermocouple)
Stabilization
Starting time: 12: 33 PM
End Time: 01: 55 PM
Stabilization Time: Approximately 1 hour and 20 minutes
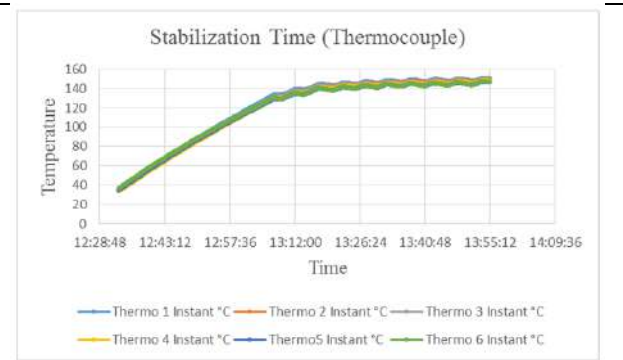
Figure 4: Stabilization (Thermocouple)

Figure 5: Empty chamber heat distribution study (Thermocouple) **2 hours loaded chamber heat distribution study (RTD)**	**Figure 6: Loaded chamber heat distribution study (RTD)** **2 hours loaded chamber heat distribution study (Thermocouple)**
Figure 7: Loaded chamber heat distribution study (Thermocouple) **90 seconds Door open and recovery study (RTD)** Study start time: 03: 52 PM Study end time: 04: 10 PM Time taken for all sensors to go out of limit: 2 Minutes (120 Seconds) Time taken for recovery: Approximately 15 Minutes (14 minutes and 45 Seconds)	**Figure 8: Door open and recovery study (RTD)** **90 seconds Door open and recovery study (Thermocouple)** Study start time: 04: 55 PM Study end time: 05: 10 PM Time taken for all sensors to go out of limit: 2 Minutes (120 Seconds) Time taken for recovery: Approximately 8 Minutes (7 minutes and 20 Seconds)
Figure 9: Door open and recovery study (Thermocouple) **10 minutes power failure recovery study (RTD)** **S**tudy start time: 03: 15 PM Study end time: 03: 45 PM Time taken for recovery: 17 minutes	**Figure 10: Power failure recovery study (RTD)** **10 minutes power failure recovery study (Thermocouple)** Study starting time: 04: 35 PM Study ending time: 04: 51 PM Time taken for recovery: 6 minutes and 40 seconds

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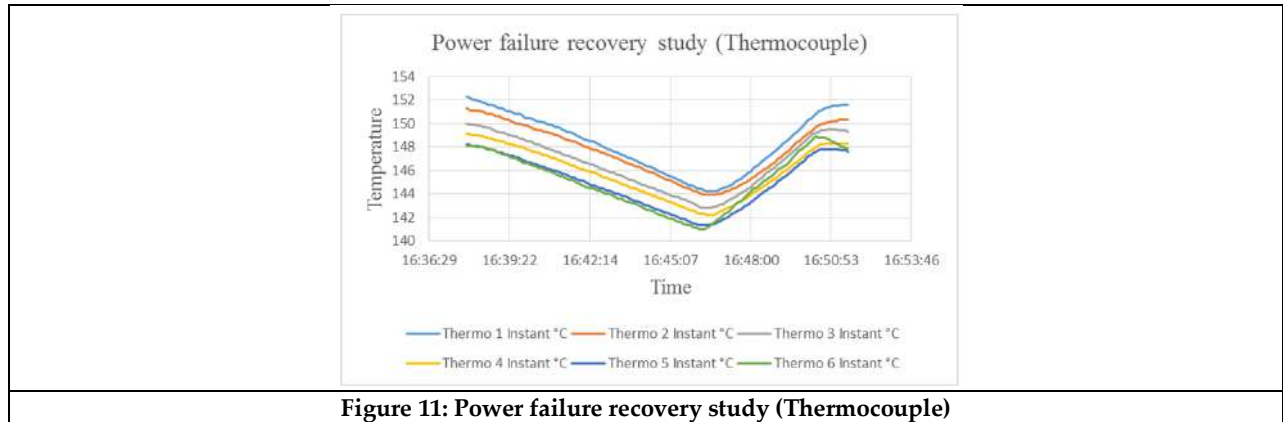
Figure 11: Power failure recovery study (Thermocouple)

RESEARCH ARTICLE

Distribution of Chronotype among Young Adulthood as Function of Gender and Age

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ABSTRACT

The aim of the present study was to investigate chronotype preference in young adulthood as function of gender and age. A random sample of 500college students, consisting of 214 males and 286 females, participated in the study. They were divided into four clusters on the basis of their age, i.e. 18 – 20 y, 21 – 23 y, 24 – 26 y, and 27 – 29 y. Each student was classified either as morning type (MT) or evening type (ET) or intermediate type (IT) on the basis of his/her performance on a Horn and Östeberg test, morningness – eveningness questionnaire (MEQ). Prevalence of MT, IT and ET in the studied population was 10.4% (52), 74.4% (374) and 14.8% (74), respectively. The males and females differed i.e. the former group had a lower score as compared with the latter group. The prevalence of IT was the highest in the studied population, irrespective of gender and age. While this observation was in agreement with those reported earlier, other components of our results differed dramatically in that the percentage of evening trait was higher (14.8%) and that of morning trait (10.4%) was considerably lower. Further, a statistically significant relationship was discovered between age and chronotype (p< 0.01). In a nutshell, although the results of the present study corroborate with the most of the earlier findings in terms of dominancy of intermediate trait, it does not support those identical reports where the prevalence of chronotype is concerned. Findings reported in this paper definitely open up new avenues for future research.

Keywords: Chronotype, young adulthood, morning type, intermediate types, evening types,

INTRODUCTION

Human behaviour exhibit circadian rhythm and is known to be overseen by the individual chronotypes (Pati et al. 2001). Documentation of chronotypes is a significant concern, since an individual's efficiency plays a substantial role

in the daily work-life timings (Roenneberg et al 2007). The chronotype of a human being has been most expansively examined (Achari et al 1997; Pati et al. 2001; Lima et al. 2002; Natale and Danesi 2002; Achari et al 2012; Achari, 2021). The behavioural outcome with the purpose of defining the degree of individuals rise early in the morning and sleep earlyknown as morning active (MT) or wake up late as well as sleep late known as evening type (ET). The individuals, who are neither morning type (MT) nor evening type (ET), are considered as intermediate type (IT).Most of the authors express behavioural typology of inter-individual differences in circadian rhythms characteristics in the human population (Natale and Danesi 2002; Hidalgo et al. 2003; Li et al.2020). Phases of the circadian rhythms in many physiological, biochemical and psychological variables occur earlier and earlier in the day in MT individuals as compared with their ET counterparts as suggested by authors (Kerkhof 1985; Achari et al 1997; Pati et al. 2001; Achari et al 2012).

The chronotype differs among diverse personalities and is reliant on on numerous factors like gender, age, and genetics (Griffin et al 1999; Scott et al 2008; Liu et al 2014).As observed the relationship between age and chronotype the inferences are still not reliable. Authors stated that the people of grater age tend to show more morning types while younger individuals exhibit more evening types (Hur et al.1998, Gau and Snoog 2003; Fischer et al 2017;Ghose et al 2019). However, it has not yet been confirmed if age-associated morning type could be imputed to the parallel changes in the circadian oscillators located in the supra chiasmic nucleus or to the changes in work and/or domestic environment (Carrier et al. 1997). Moreover, it is also documented that males in the younger age exhibit more evening types than that of their counterparts females. However, in some studies no significant differences in the distribution of chronotype score were found between females and males (Posey and Ford 1981; Ishihara et al. 1988; Neubouer 1992; Achari et al 1997: Achari et al 2012). In disparity, many studies propose that females tend to score significantly more towards morningness than males (Chelminski et al. 1997; Ghose et al 2019).Scientist still has difficulty in giving an exhaustive explanation for the relationship between gender, age and chronotype preference. Therefore, an unanimity of opinion is yet to emerge. Therefore we consider that gender differences in chronotype preference should be studied further. The present study aimed to ascertain the prevalence of chronotype among college student of central, India, with special reference to gender and age.

MATERIALS & METHODS

Subjects
Five hundred college students of central India were participated in this study, which consist of 214 males and 286 females. The subjects were aged from 18 to 28 years (19.5 median). They were further classified into four groups on the basis of age, viz; 18-20y, 21-23y, 24-26y and 27-29y.

Horne and Östeberg Test
All participants were subjected to personal interview with a set of Morningness-Eveningness Questionnaires. The test was based upon a score scale ranging from 10 to 28. The subjects were identified with either morning (MT) or intermediate (IT) or evening trait (ET) depending upon the total score they achieved of 7-14 or 15-22 or 23-30, respectively, on the Horne and Östeberg test (Horneand Östeberg, 1976).

Procedure
The questionnaire of Horne and Östeberg was administered to each and every subject at the commencement of a schedule instruction. After a brief foreword to the nature of the research and ethical requirements for confidentiality, voluntary participation and informed consent, they were instructed to participate. Most of the questionnaires were completed and returned to the researchers at the end of the lecture with personal discussion. The remainder were collected within a maximum lag of oneweek with personal discussion.

Statistical analysis
Data were stored in the form of records in a database file. All data were analysed with the help of software, namely SPPC. The Chi-square test and ANOVA were employed.

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RESULTS

Out of five hundred subjects, 52 (10.4%) were morning, 374 (74.8%) were intermediate and 74 (14.8%) were evening type (Figure 1). The MEQ score range of total subjects was 10- 28 (Median18). However, the range for males was 10-28 (Median20) and females it was 10-28 (Median19). Chi-square test indicated that there was no significant relationship between gender and circadian typology but there was significant relationship between gender and age groups (Table 1). The percentage of morning type males (11.2%) was greater than that of females (9.8%), while it was almost same for evening type and intermediate type distribution (Table 1). Results also depict that the prevalence of intermediate traits was the highest in both genders. The results of the Chi-square test indicated a statistically significant relationship between age and chronotype. Further each age group was interpreted gender-wise and it was revealed that the age effect was independent of gender (Figure 2).

DISCUSSION

The results of the present study exhibited predominantly more intermediate type traits (74.8%) among the college adulthood, irrespective of gender and age. Authors also reported that the morning types were low (10.4%) than that of evening types (14.8%). Nonetheless, the results of the present study contradict the results of an earlier study conducted by authors in whom evening types were extremely lower (1.93%) than that of morning types (Achari and Pati 2007). These authors worked on the adolescent age group (8y to 23 y) while we are worked on adulthood (18y to 28y), it seems that when human being switch over to adolescent to adulthood the traits use to amend slightly from evening types to morning types. Prior to present study authors testified the same occurrence in adult population (Natale and Danesi 2002). In the present study although the females had higher mean score that that of their counter parts, the results of the chi square test did not reveal a statistically significant association between the gender and chronotype. However, our study results based on gender wise distribution are in agreement with those reported earlier (Chelminsky et al 1997; Natale and Adan 1999).

Further, a significant relationship was observed between different age groups and chronotype. The intermediate types were predominantly more than that of their counter parts. However, a significant difference could be validated within different age group, when participants were classified into four age groups. It seems that age plays significant role in the establishment of chronotype irrespective to clock gene (Caci et al 2005). Correspondingly, chronotype trait has been reported to be independent to *hPer1* and *hTim* in adult and adulthood (Katzenberg et al 1999; Pedrazzoli et al 2000). Prior to this statement authors emphasized the beginning of adolescent age established evening types (Caci et al 2005; Fischer et al 2017;Ghose et al 2019). It could be attributed to hormonal fluctuation during adolescent female tend to exhibit evening types (Takeuchi et al 2001; Fischer et al 2017;Ghose et al 2019). Dissection of chronotype study with reference to age might produce appreciated conclusion. In a nutshell, although the results of the present study substantiate with the earlier findings in term of predominant intermediate types than that of morning type and evening type. The present study validate with the finding that the distribution of chronotype exhibit partially Gaussian patterns. This study would be evidence of independent factor age and open up new path for further research.

ACKNOWLEDGEMENTS

DECLARATION

The authors report no conflicts of interest. The authors alone are responsible for the content and field work. The entire work is self-funded by author.

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Table 1 Distribution of chronotype as function of gender			
Chronotype	**Gender**	**% of population (N)**	**χ^2–Value; df; p- value**
Morning Type	Male	11.2% (24)	0.276; 2;0.871[ns]
	Female	9.8% (28)	
Intermediate Type	Male	74.3% (159)	
	Female	75.2% (215)	
Evening Type	Male	14.5% (31)	
	Female	15.0% (43)	

[ns]Non significant

Figure 1: Distribution of chronotype among college students of India irrespective of age and gender(MT =morning type, IT = intermediate type (ET = evening type)	Figure 2: Distribution of chronotype as function of age (MT =morning type, IT = intermediate type (ET = evening type)

RESEARCH ARTICLE
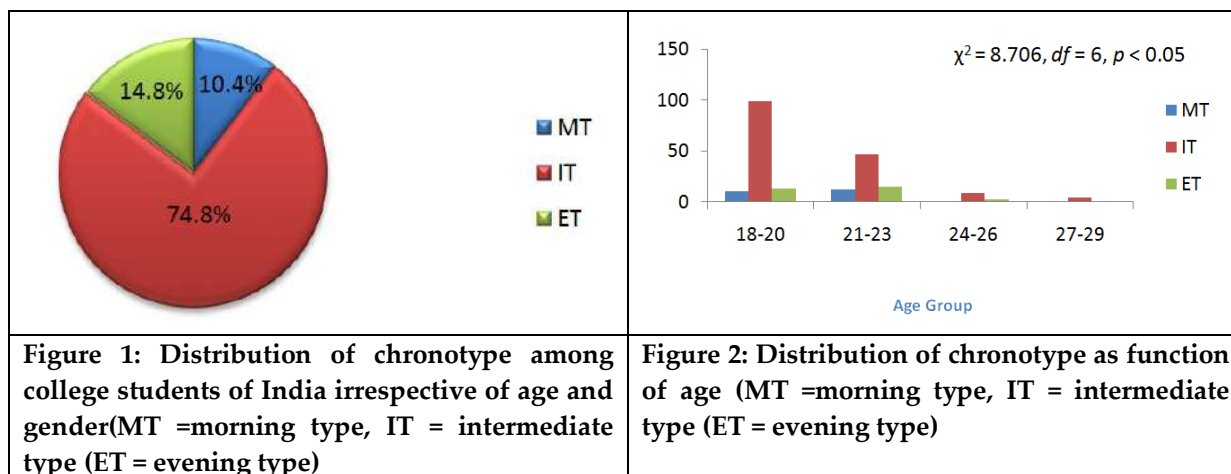
Analysis of sensor network in Different sectors

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ABSTRACT

Networks that collect data about the physical phenomenon of entourage without the usage of wires but with the usage of interconnected sensor nodes are known as Wireless sensor networks. They are useful and provide many applications and give us the comfort of smart economic life. It is helpful to figure out the noise and atmospheric pollution. We can also check the heart rate and pulses of the patient without the usage of wires. Sensors are used in agriculture for monitoring plants, measuring temperature, irrigation system, measuring water supply etc. all from a far-flung through the sensors (camera). Sensors are used in the bakery for cutting the shape, mixing dough and in baking mainly to keep a check on the temperature. Sensors are used in hospitals to check the blood pressure of a person, as it detects physical phenomenon. It is also used in automobiles mainly in cars and scooters to keep a check on acceleration, amount of fuel consumption, identify car pressure and health motoring system of the driver.

Keywords: sensor, health, phenomenon, sector, network, usage

INTRODUCTION

Wireless Sensor Networks (WSNs) enjoy great benefits due to their low-cost, small-scale factor, smart sensor nodes. Not only can they be employed in unmanageable and dangerous areas of interest, for observing or managing the region, but they can also be deployed to automate mundane tasks. At the beginning sensory units were expensive and lacked the computational and communicational capabilities of current smart sensor nodes, which can now sense, process, store, and forward data, all being powered by a battery. Myriad applications exist that leverage WSNs as low-cost solutions for observing the habitat and environment, from army and public surveillance and target detection and tracking applications, to precision farming and agriculture, patient monitoring in hospitals, residential applications like energy management, for safety and efficiency in vehicular networks to outer space explorations.

The i-Button: is a computer chip enclosed in a 16mm thick stainless steel. Because of this special and durable container, up-to-date information can travel with a person or object anywhere they go. The iButton scale of dataloggers is extremely small and rugged, offering excellent value for money. The iButtons' unique size makes it possible to log temperature and humidity data in locations that were previously unthinkable.

Simputer: Agri-information: The Sirnputer is a low cost portable keyboard less handheld device with a (320x240) LCD panel which is touch enabled; a speaker, microphone and a few keys and soft-keyboard by which the benefits of IT can reach the common man. It has a special role in the third world because it ensures that illiteracy is longer a barrier to handling a computer. Simputer may be split by a conmmnity at either Panchayath office, community center, SHGs etc. The effect of this feature coupled with the rich. Integration of the Simputer can be dramatic. Hence, Simputer, is not as an end product but as an evolving platform for social change. This paper highlights the applications of Simputer in diverse sectors such as micro banking, large data collection, and agricultural information and as a school laboratory etc.

Greenhouse Monitoring: WSNs used to control the temperature and humidity levels in greenhouses as shown in figure 4. Fall of temperature or humidity below a piece of specific level. Information transmitted by the node to the base station which can send a notification to the greenhouse manager.WSNs is easy to move Ideal for this kind of application.

Precision Agriculture: Precision Precision agriculture employs data from multiple sources to enhance crop yields and increase the cost-effectiveness of crop management strategies including fertilizer inputs, irrigation management, and pesticide application. Distant sensing has been promoted as a key source of information available in support of precision agriculture for decades, but adoption has been slow for a variety of reasons. Latest technological developments, specifically the availability of high-resolution satellite imagery, and more recently, rapid development of unmanned aerial vehicle technology (UAV), suggest that adoption of remote-sensing data sources in precision agriculture is likely to rapidly.

Analysis of Soil Moisture: Analyzing the soil moisture sensor due to the effects of the electrode size and number of ground electrode.

Applications:
- The best way to solve the agricultural problems related to farming resources optimization, decision making support, and monitoring of land.
- Increasing production efficiency, improves product quality, improves the efficiency of crop chemical use Conserves energy, and protects the entourage.
- To help protect their crops from frost damage, there are temperature sensors being used. The temperature motes will help Farms target specific plants for frost-control measures, such as misting the plants with water.
- Analyzing long-term data from the sensors, they'll also be able to harvest crops more productively and cut their use of pesticides and fungicides.
- Reducing the level of the pesticides.
- While ensuring a best production.
- Determining the disease development during the growing season.

WSN Applications: healthcare: Large-scale in-field medical and behavioral studies: Body-worn sensors together with sensor-equipped Internet-connected smartphones have begun to massively used by medical and public health research studies by enabling behavioral and physiological data to be continually collected from a large number of distributed subjects as they lead their day to day lives. With their capacity to provide insight into subject states that cannot be replicated in controlled clinical and laboratory settings and that cannot be verified from computer-assisted retrospective self-report methods, such sensing systems are becoming critical to medical, psychological, and behavioral research.

Indeed, a major goal of the exposure biology program under the National Institute of Health (NIH) Genes and Environment Initiative (GEI) is to develop such field-deployable sensing tools to quantify exposures to the environment (e.g., psychosocial stress, addiction, toxicants, diet, and physical activity) objectively, automatically, and for days at a time in the participants' natural environments. Researchers, both within and elsewhere of GEI program also have recognized the utility of such sensing in making measurements for longitudinal survey ranging from the scale of individuals to large populations.

Robotics: Nowadays many applications use WSNs and robots. Robots can cooperate and combat some of the major problems of WSNs, such as sensor node mobility, node redeployment, traveling salesman, etc. here are some typical WSN applications presented below. A robotic navigation method provides road maps for the robot to traverse. It uses a WSN with sensors, designed to provide detailed maps of their working areas. Specifically a map is constructed by each sensor, based on the attainable area sensed. One large map is created by combining small sensor maps. Once the road maps are generated, it is used by local citizens as GPS (global positioning system). The robot then checks the status of the traffic and provides the best road in the network. If an area becomes dangerous for the robot, the network can change the route of the road map, and remove this dangerous area from the list of available paths. One of the hardest tasks concerning WSNs is the maintenance of a projected network. A combination of WSNs and a robotics network servicing system, named Randomized Robot-assisted Relocation of Static Sensors (R3S2) was developed. In R3S2, robots move around a network that is contained within a virtual grid. The robot takes to the least recently visited grid point, searching for sensing holes in the network. if an area that is not being covered by a sensor is detected, the robot will find a node that has overlapping coverage with other nodes and move it to the uncovered area. In advance, if the robot discovers redundant sensors, it will move the nodes to cover a greater area. A different approach, where a mobile robot is used to transfer data from a widespread network between nodes that are out of reach from one another for various reasons. This is the so-called traveling salesman problem (TSP) within a WSN. By having a robot traveling among nodes that are out of each other's wireless communication range, it allows the network to be widespread while also saving power by using the robot for data mining

CONCLUSION

Advantages for various domains of human activity. Due to the evolution of technology both the capabilities of sensor nodes will be manufactured at a lower cost, i.e. the product will low cost and available to all. This is the main reason for WSN success. In this article, the utilization of WSNs in specific domains, namely agriculture, health, industrial, and urban, was examined through the investigation of similar typical examples, both novel, and known form. From this examination, it became clear that the usage of WSNs not only provides a number of advantages in specific fields when compared against the relative means and methods that are traditionally being under use.

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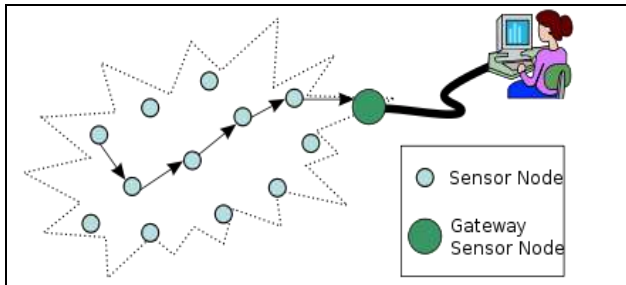
Examples of Applications of WSN in Agriculture	**Figure 01: view of iButton**
Figure 02: view of Simputer	**Figure 03: view of Greenhouse Monitoring**
Figure 04: Model of Precision Agriculture	**Figure 05: A Test setup for the FEF Soil**
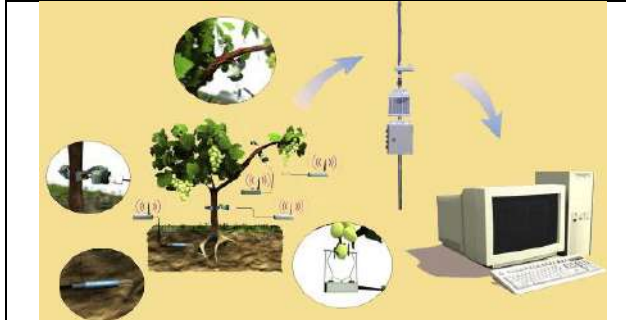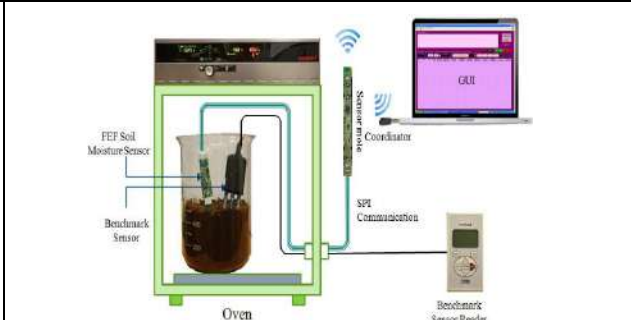
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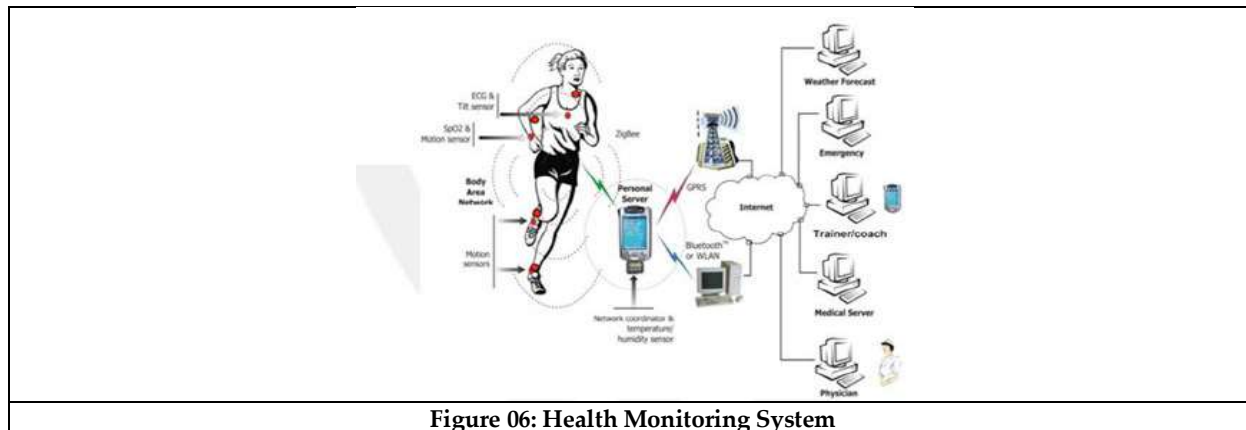
Figure 06: Health Monitoring System

RESEARCH ARTICLE

Crop Yield and Locust Prediction using Machine Learning with Augmented Reality Smart Cards. (Khet-E System)

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ABSTRACT

Machine learning is one of the emerging technologies in Artificial intelligence that provides the ability to learn, infer, re-correct and relearn from the experience gained. The robustness of Machine learning and Deep learning can be highly utilized in any domain especially for agriculture related production system. The precision and the strength of these algorithms helps the farmers to take appropriate decisions and supportive actions. The research work has concentrated on vital sections of agriculture that has a drastic impact on farming and farmers. This paper gives a detailed explanation on five categories. They are (a) crop yield prediction to rise the nourishment of the crops using Machine learning algorithm to check the nutrients of the crops, soil fertility, (b) provides suggestions to grow crops reliant to the geographic location and the amount of rainfall received, (c) fertilizer recommendation with respect to the species and crop recommendation, (d) disease prediction in crops and livestock (e) Locust attack prediction. The prediction and recommendations for the above-mentioned analysis were provided by Machine learning and Deep Learning algorithms. The research work has been further extended to develop a smart card using Augmented Reality that helps the legitimate farmers to communicate through a common forum, and to have a B2C model sales and purchase support. Application of artificial intelligence in farming and farm management provides enriched experience and recommendations to the farmers that helps them to take appropriate actions without wasting time, resources and energy.

Keywords: Artificial Intelligence, Augmented Reality, Machine learning, Deep learning, B2C, Convolutional neural networks

INTRODUCTION

Machine learning is a critical decision support tool for estimating crop yields. This includes assisting with crop selection and suggestions during the growth season of the crop. Crop yields are difficult to estimate since a variety of

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factors influence them, including crop genotype, environmental factors, management strategies, and their interactions. This paper uses convolutional neural networks (CNNs)to create a framework with deep learningfor yield predictions based on ecological information and management methods. The proposed CNN model was integrated with other traditional approaches like Random Forest (RF) and Deep Fully Connected Neural Networks to estimate agricultural output (DFNN). The precision and the strength of these algorithms help the farmers to take appropriate decisions and supportive actions. Technologies like Augmented Reality (AR) and Virtual Reality (VR) have exploded in popularity recently. There have been several hardware devices and techniques introduced accordingly. AR (augmented reality) is a technology that allows virtual objects to interact with their environment in real time. However, as AR became available on smart phones, it allowed for widespread adoption of AR applications in fields like education, manufacturing, military, and health, among others, by presenting users with information on designated targets. It's also one of Industry 4.0's most essential technologies. AR systems have already progressed to the point where their efficacy has been widely established in the educational sphere and assists real farmers in communicating through a common venue.

A subset of machine learning is deep learning that allows computers to acquire knowledge by doing, much like humans do. Deep learning (DL) improves recognition accuracy beyond what has ever been achieved previously. A computer model learns to execute categorization tasks directly from pictures, text, or sound in deep learning. Deep learning representations might achieve state-of-the-art accuracy, sometimes even outperforming humans. Multilayer neural network topologies and a large amount of labeled data are enabled to train models. Neural network architectures are the base for the deep learning approaches. Combination of both the models re mentioned as Deep neural networks (DNN). DL is trained using large dimensions of categorized data and neural network topologies supposed to observe the structures straight from the information without human feature extraction necessity. Convolutional neural networks (CNN or ConvNet) [2] is the most prevalent type of DNN. A CNN combines learned characteristics with incoming input using 2D convolutional layers, enabling it perfect for processing 2D data like images.

CNN eliminates the necessity for feature extraction manually, enables the user not figure out what features are utilized to classify photos. CNN directly extracts the attributes from photographs. The necessary properties are not pre-trained; rather, they are found when the network is trained on a batch of images. The self-learned feature extraction in DL model provides extremely accurate computer vision classification applications.

Proposed System

The objective of this project is to give a higher yield and more sustainable farming for farmers, as well as to assist malnourished animals in receiving nutritious fodder and to establish a local online market where farmers may purchase/sell their crop, rent or buy farm machinery. The review recognizes that farmers may find it difficult to utilize websites, therefore a WhatsApp Bot has been developed with the functionality of webpages to reach a wider audience.

Working of the Proposed System

Crop Recommendation - The user may submit soil data (Nitrogen, Phosphorus, Potassium concentrations, pH value, Rainfall in mm, State and City) and therefore the program will recommend which crop the user should produce. The system utilizes an open weather API to authenticate the temperature, wetness, and humidity for the desired location automatically.

Fertilizer Recommendation - The user may input soil data and therefore the type of crop they are growing into the fertilizer recommendation program, and also the application will forecast what the soil lacks or has a way over, and can make recommendations appropriately.

Disease Prediction - within the illness prediction application, the user uploads a picture of a crop infected with the disease, and therefore the model predicts the disease, explains the rationale, and suggests a remedy.

Huang et al. [2]Introduced Dense Net, a new architecture that takes use of the effects of shortcut connections by connecting all layers directly. ResNets use a network with residual blocks, where each layer feeds into the subsequent layer and straight into the layers 2–3 hops away, rather than typical neural networks, where each layer feeds into the subsequent layer. This reduces over-fitting (a situation when validation loss stops decreasing at a point and then keeps increasing while training loss still decreases). This also allows us to train deep neural networks while avoiding the vanishing gradient problem. An example of a basic residual block is as follows:

The overall framework over stochastic depth training is demonstrated in the figure below

The picture above depicts the entire structure for stochastic depth training. During the training process, deactiavtion is possible in all layers.

Add Ons

1. Soil Classification –The system produces a streamlit-based soil classification add-on that allows you to categorize soil using a picture of the soil.

2. Nursery Locator –The proposed system has a feature that allows the farmer to find a nursery near you that sells the crops and fertilizers you require.

3. Utilize the WhatsApp Bot Feature –The proposed systemhas a user friendly button that connects to theWhatsApp bot.

Twilio-Based Whatsapp Bot

The bot is designed in a manner that it first learns the language the farmer use to contact it and then utilizes that language to ask questions and offer responses. There are four options available to the bot.

1. Crop Suggestion - In the crop recommendation option, the user enters their soil data (Nitrogen, Phosphorus, Potassium concentrations, pH value, Rainfall in mm, State, and City) and the bot recommends which crop to produce.

2. Fertilizer Suggestion - The user may input soil information and the category of crop they are growing under the fertilizer recommendation option, and the bot will forecast what the soil lacks or has an excess of, and will make recommendations appropriately.

3. Disease Prediction - With the disease prediction function, the user may upload an image [3] of a crop infected with an ailment, and the bot will diagnose the disease, explain the reason, and suggest a solution.

4. Change Language - If a user wishes to change the language, they can send the message Change Language = Lang>, and the bot will do so.

The Locust Tracker Map

This function is incredibly useful and beneficial to farmers since it records the locusts' movements and notifies the farmers in the direction in which they are going, allowing farmers to securely maintain track of their harvests.

1.   The card for Augmented Reality-
2.   When the AR card is scanned, the user is sent to anyone of four symbols that represent valued services:
3.   The webpage of Khet E.
4.   Crop Recommendation, Fertilizer Recommendation, and Disease Prediction are all included in this module.
5.   The bot for WhatsApp.
6.   The locust tracker is a map that shows where the locusts are.

EXPERIMENTAL RESULT

This section explains the result experimental results and the working machine learning model. The screenshots are from the proposed Khet-E system for smart card services, crop recommendation depending on soil types, disease prediction and fertilizer recommendation has provided in the following section.

Figure 7 depicts the various algorithms utilized and their accuracy.

As illustrated in Figure8, the smart card displays the functionality of applications in one forum utilizing augmented reality. Figure 9 illustrates how to determine the soil type and the best crop for it. Depending on the soil data entered the user has suggestions for what type of crops that could be cultivated as depicted in Figure 10. The predicted crop suggestion after the given soil data input is shown in Figure 11. The user may input soil data and the sort of crop they are cultivating. The user is given an abstract of what their soil is lacking and is suggested the recommended fertilizer add-ons for better yield as sown in Figure 12. With the disease prediction function, the user may upload an image of a crop infected with an ailment in order to know about the disease that the plant is infected with as depicted in Figure 12. The ML model is depicted in Figure 12.1, which predicts the disease, explains the reasons, and offers a treatment. Incredibly useful and beneficial to farmers since it records the locust's movements and notifies the farmers to the direction in which they are going, allowing farmers to securely maintain track of their harvests as depicted in figure 13. The map indicating the yield and fertilizers that are put up for sale in the zone the user wishes to purchase from as depicted in Figure 14. Figure 15 shows the Homepage of the agricultural goods purchasing and selling business-to-consumer industry. Farmers may choose from a diversity of options on the B2C platform as depicted in figure 15.1. Registration for a supplier to participate in the selling process as depited in figure 16. The admin dashboard, shown in Figure 17, displays all of the information about the website for buying and selling. The website as given in Figure17.1 provide privileges to rate and review the supplier.

Twilio WhatsApp Bot:
The user enters their soil data (Nitrogen, Phosphorus, Potassium concentrations, pH value, Rainfall in mm, State, and City) and the bot suggests which crop to grow as shown in Figure 18. Under the fertilizer recommendation option, the user may input soil data and the type of crop they're growing, and the bot will anticipate what the soil lacks or has an abundance of, and offer recommendations accordingly as depicted in Figure 19. The user may provide a photograph of a crop affected with the illness as shown in Figure 20, and the bot will identify it, explain why, and recommend a solution as shown in Figure 20.1

CONCLUSION

The Khet-E system is a multifunctional system that alleviates the different issues experienced by farmers in this contemporary day, where things like crop yield and quality are efficiently increased to their peak with the assistance of the Machine learning portion of the proposed system's services. Buying and selling their harvest or farm machinery eliminates the need for a third-party intermediary, and the safety of the crops is monitored with the use of additional services such as the locust tracker map, disease prediction, fertilizer advice, and so on. These services assist the farming community in time management, productivity, and a consistent high production, which is advantageous to both consumers and sellers. The khet-E system is simple to use, with user-friendly APIs, and it is also multilingual, making it more resilient for usage in nations like India, where the population is heterogeneous.

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Datasets:
Crop Recommendation Dataset | Kaggle
New Plant Diseases Dataset | Kaggle

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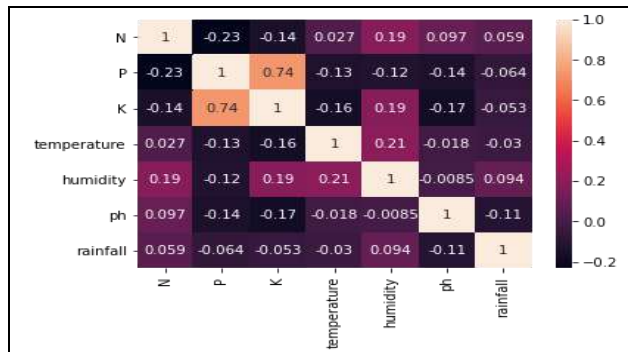
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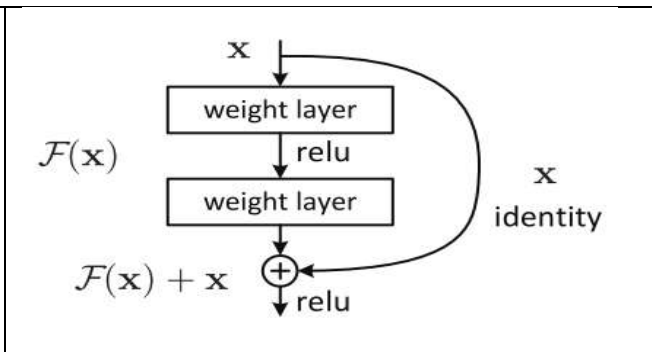
Figure2: Residual learning: a building block [1]
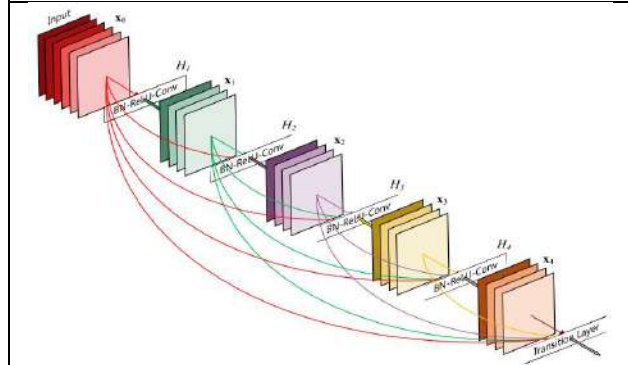
Figure3: Densely connected CNN [1]
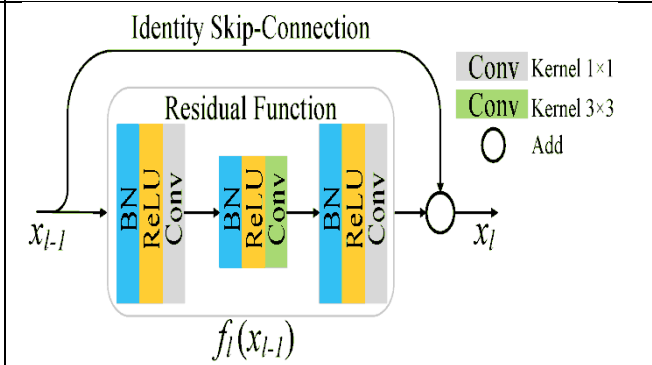
Figure4: Residual Block code implementation[2]
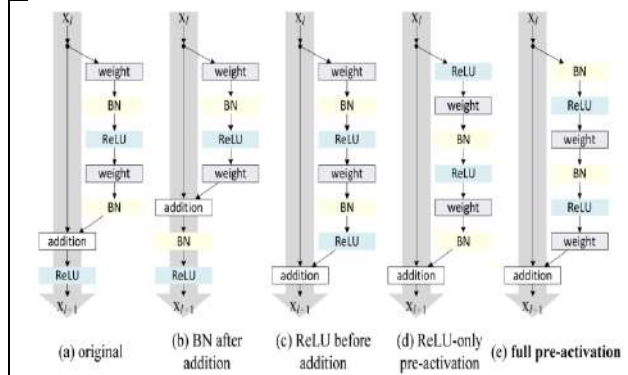
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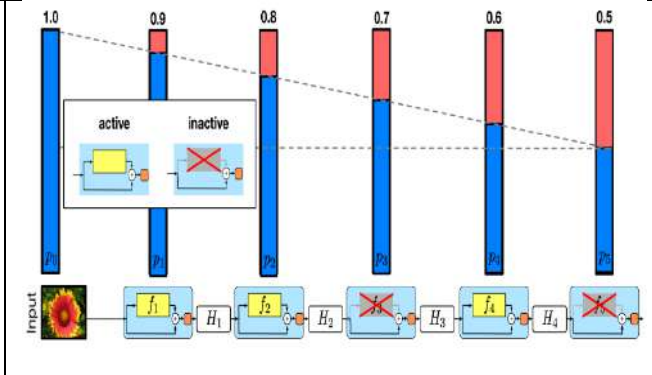
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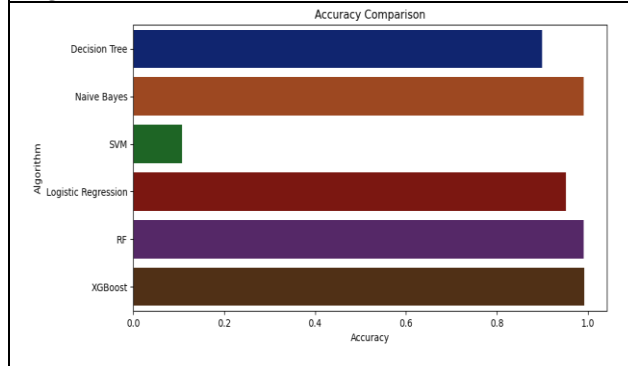
Figure7: Accuracy Comparison vs algorithm

Figure8: Augmented Reality Card

Figure9: Soil classification ML	**Figure10: Crop prediction ML**
Figure 10.1: The outcome of crop prediction	**Figure 11:Fertilizer Suggestion**
Figure11.1:Fertilizer Suggestion	**Figure12: Disease Prediction**
Figure 12.1 Prediction Model	**Figure 13: The locust tracker map**
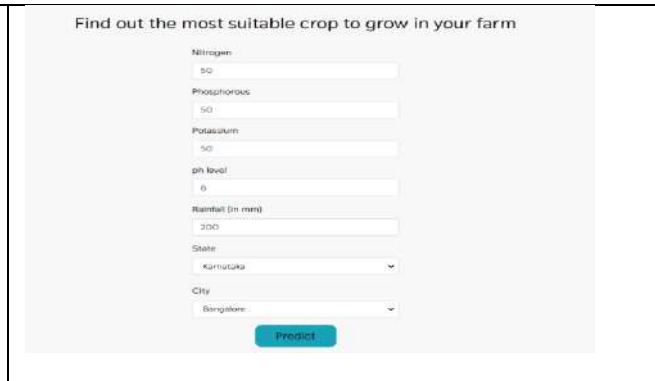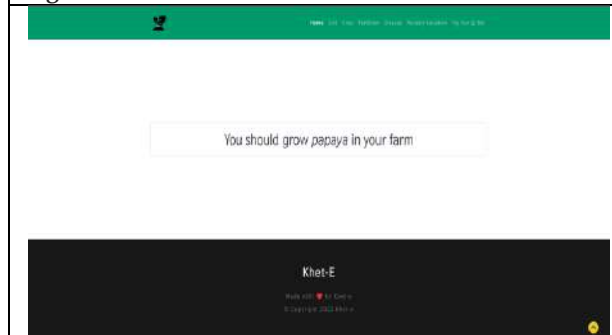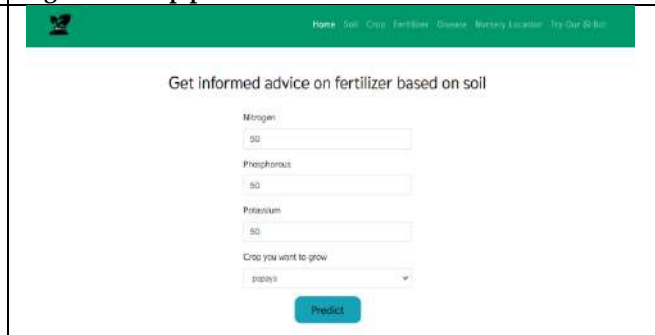
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Figure14: Nursery Locator

Figure 15: The Khet E website
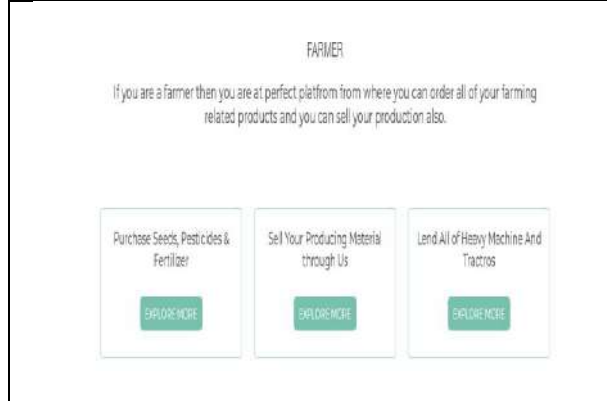
Figure 15.1: Options offered for farmers
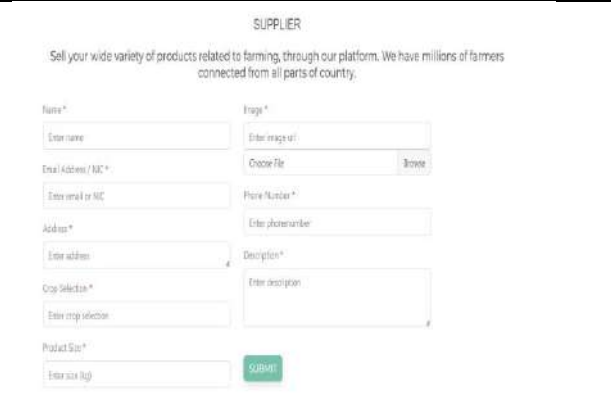
Figure 16: Registration for supplier

Figure 17: Admin Dashboard
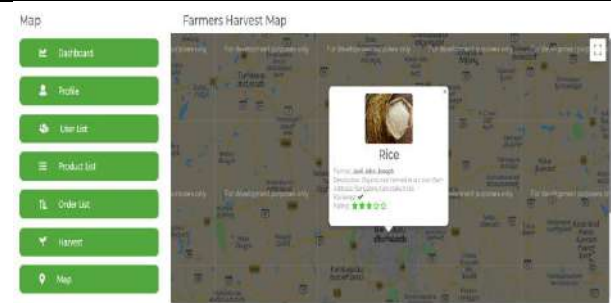
Figure 17.1: Rating and review of supplier
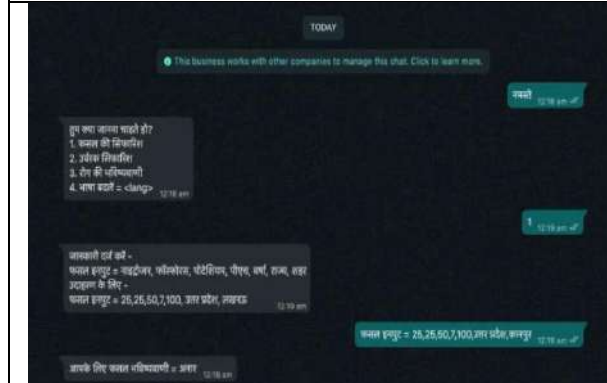
Figure 18: Crop Suggestion
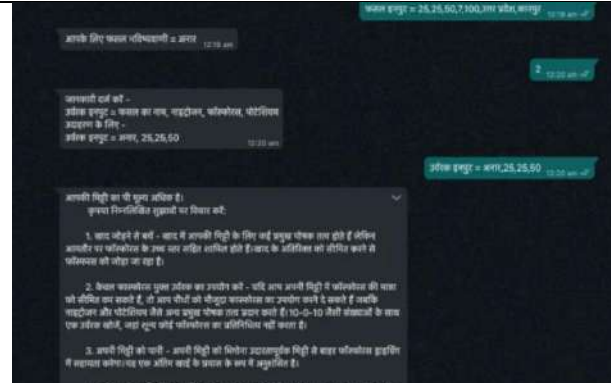
Figure 19: Fertilizer Suggestion
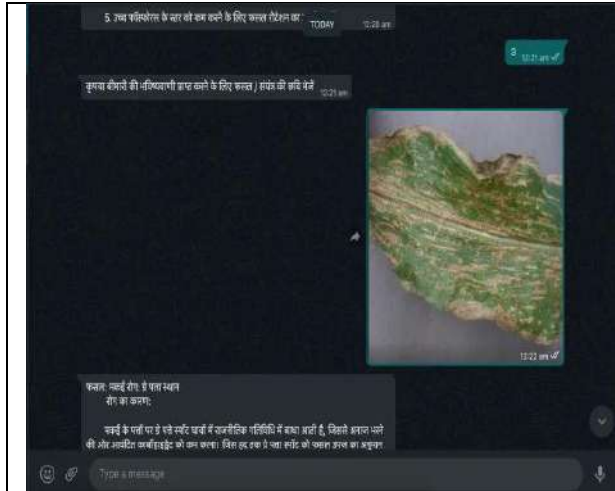
Figure 20: Disease Prediction	**Figure 20.1: Recommended Solution for a specific disease prediction**
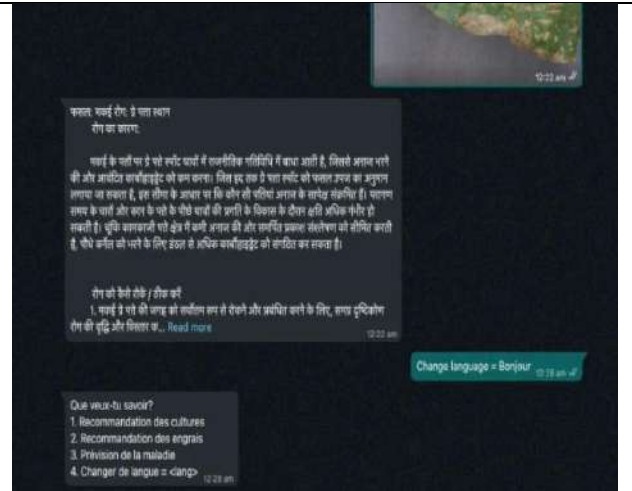
RESEARCH ARTICLE

High Peak to Low Peak Analysis using Emitted Sound Signal in Tool Wear Monitoring

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ABSTRACT

Industrialized manufacturing requires optimization methods in fabrication activities as well as tool wear monitoring. The unmonitored tool is left may injure the affected product condition is exhausted additional bearable levels otherwise wrecked or cracked. An innovative narrative technique recommended during this job utilizes no further standard calculations or complicated statistical transformations. In this paper peak to peak study the high peak and low peak wavelength and frequency analysis results in instead of the tool flank wear degree. Continuous examination through the proposed method on different statistics sets of three categories state fresh, slight as well as severe resulted in important threshold distances on together high peak and low peak.

Keywords: Tool Flank Wear; Tool condition monitoring; Microphone; Tool sound;

INTRODUCTION

Automation is involved in almost known human intervention in any step of manufacturing process. In CNC job work the production process greatly depends on the product manufactured using the condition of tool in the job work should be an authentic fine quality product. In the turning process of automation as well as conditional monitoring of an effective and active tool, the drilling and milling process requires a mechanism that job may hamper the sequence of operation. The condition of tool wear is regularly monitored without disturbing the machining process in order to make perfection of the product and tool wear condition sure. Many conservative methods of putting in sensors and camera's to deal with the issues have been suggested but involve so many factors together with condition of machines. The process variables in case force of cutting, hearing emission sense, current of spindle motor, heat, roughness, vibration, from the surface or relationship between the tool wear value are observed is the indirect method of tool condition monitoring by adding values for some required parameter. Sound signal are introverting

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the information from evaluated feed motor current, feed cutting force are the parameters of Alonso et al. recommended a guessing method of tool flank wear a ANN. The directly proportional correlation between the ceiling extend of vibration with the rising tool wear was established by Sadettin *et al* (2007). Through cutting method the shaped audible sound is used like device in support of monitoring for the tool wear commencing the research of Alonso F.J et al. and Ming *et al* (2005) commencing the converting process to expect the tool flank wear swiftly via dynamic assessment group technique through a portion of discharged noise by Samraj *et al*(2011). The distinctive signal is handled in the form of linear and not permanently immobile; otherwise besides the consequence deriving Fourier spectrum could generate drastically lesser physical intelligence was recommended by Peng *et al*(2005).

Fourier transformation leads the global properties of the signal rather than local properties according to the trend of Huang et al. Since it recruits a coil or twist integral through that the signal breaks down in the form of sine function as well as cosine function with the purpose of covers the entire data extent homogeneously. The transformation of wavelength survey of frequency in time is used toward fabricate time and frequency in sequence of the single dimensional signal obtain uninterrupted aligning in the direction of time frequency plane for two dimensional. Sick (2002) through experienced research in excess of decade taking place monitoring through online of unplanned tool wear by perverted neural network come to an end that it is workable to differentiate in the tool wear parameter with neural network. An intellectual capacity for advanced analysis of emitted sounds for entire converting procedure in the direction of estimation the tool wear condition could be sensible method with the purpose of augments several competent manufacturing through automated. The alternatives resembling images captured within videos along with other devices used for monitoring may cause expenses and erroneous predictions. This paper deals with the evaluation of wear in distinct point cutting tools by plotting the sound waves emitted during the cutting operations for analysis and the parameters calculation.

METHODOLOGY

Hardware Setup

The substantial quantity of sound is created for the duration of the turning procedure like a result of the vibration formed beginning the work-piece as well as work machine tool. The produced sound whereas process to be estimated according toward the power in addition towards the dimension of facade of make contact with to which the supplement of cutting flank wears occurred. The length of the other mixed vibrations formed environment within the tool wear estimation method. These trembling interruption be able to estranged by means of suitable filters because they appear during the fewer than average frequency ranges is null point [0] in addition to 2 kilo Hertz. Whereas the effect of the conversion procedure is predominant as well as significant on these interruptions over 2 kHz frequency measure. The level of noise pressure is calculated through condenser microphone characteristics starting such generate sound wave. The Figure 1 shows the sound wave to be record using microphone

The micro phone is commercially accessible Prepolarised Control Block 130 A24 is used during this research to record the vibration of sound. This PCB 130 A24 microphone is able to record an essential amount of noise up to 12decibels. This microphone is more appropriate in the direction of receive as well as record the sound of turning process by means of reaction toward the incidence between 20Hertz to 20 kiloHertz through the exactness of sound varies ±0.5decibels. PCB130A24 microphone be able to withstand high temperature because it is having BNC connector, subsequently the requirement for outside polarization was unconcerned, which comprise freeze concerned with charges could be implemented on peak of the backside plat's. This prepolarised control block 130 A24 microphone was broadly used within measuring sound during more than one channel machinery in addition to power measurements using sound.

The preparations and location of the PCB 130 A24 microphone is associated towards measuring are exposed in Fig. 2. Shows the recording process during the machining process. In this experiment the Aluminum material work-piece is preferred contain a width approximately 40 mm. In addition to that insert the cutting tool bit used for the period of

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this experiment. This micro phone outputs differs according to the sounds force on top of the instance discipline proportionally to the alterations made in the sound pressure taking place the time discipline. A 'Gold Wave' software is connected to the microphone which helps to perform the sound signal recording along with the modal rate of 44100 with existing PCB 130 A24 microphone;. The rate of sampling is high in addition to the output data outstanding in the direction of this elevated sampling rate is a demanding more than head for processing. The single signal having the measurement of 1 x 44100. The Experiments was intended with a small number of constant cutting parameters like cutting speed through feed rate. The emitted noise signal is recorded 20 experiments through the turning function. These recordings to be done along with three sessions of every one the three position conditions of the aluminum material tool bit. The sound of free run is recorded towards include as a bottom reference during the start of the experiment whereas the machine is freely operated exclusive of moving the work piece through the tool small piece. This exacting measurement is afterward named while no controls run. Consequently tools for assessment similar to Fresh tool, severely wear recordings in addition to slightly wear tool were also labeled since the noise waves are recorded. The experimentation as well as inspection are done through the 10X3 interleaved samples designed for each category of tool wear. The transformation of samples into its digitized from using 'wave read' function. For the future evaluation we can use the recorded digital signals in form of array.

RESULTS AND DISCUSSION

The detection of tool wear is able to be recognized by our future method exclusive of any transformations. The signal samples in use over the period of time were analyzed with high peak and low peak averages for three categories of sound signals.

Peak to Peak Analysis
The peak to peak analysis was done in two levels for the splitted data set using interleaving methods. The purpose of down sampling or interleaving is to make sure the fitness of the projected method by minimize the freight and complication. The wavelength and frequency are shows in figure 4, 5 and 6.

Covariance Analysis
The covariance analysis was done in two levels for the splitted data set. Variance refers to the spread of a data set around its mean value, while a covariance refers to the measure of the directional relationship between two random variables represented in table I. The figure 7 shows the averages of covariance analysis of fresh, slight and severe wear categories.

T- Test Analysis
The t test can be used to determine the significant difference between the means of two different data set groups, which may be related in certain features of the sound. The significant difference is 0.05.
Table II presents a significant average difference for the three categories of tool bit sounds. The average of fresh and slight significant difference is 0.544. Similarly the slight and severe significant difference is 0.4100 and fresh and severe significant difference is 0.3238 are shown in the above table.

CONCLUSION

On observing the tabulated, result our proposed technique of high peak to low peak analysis on both the data sets resulted in good differences and in proper intonations. This approach is easy and does not engage several difficult mathematical transformations or derivations. The significant difference calculated is found through t test by using 0.05 differences. So that the purposes of tool wear monitoring is ended accurate and easy.

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Table I: Covariance analysis of three categories of fresh, slight and severe			
Function	**Fresh AL**	**Slight AL**	**Severe AL**
F1	0.3138	0.3436	0.2121
F2	0.3956	0.3402	0.2146
F3	0.5127	0.3392	0.2285
F4	0.4226	0.3386	0.2084
F5	0.4386	0.3621	0.2388
F6	0.4615	0.3355	0.2225
F7	0.4980	0.3888	0.2250
F8	0.5536	0.5220	0.2217
Average	**0.44955**	**0.37125**	**0.22145**

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Table II T-Test analysis of three categories of fresh, slight and severe tool bit sound						
Function	Fresh	Slight	Slight	Severe	Fresh	Severe
F1	0.5535		0.4151			0.2816
F2	0.5965		0.4938			0.3785
F3	0.4395		0.3198			0.3174
F4	0.4959		0.4448			0.3349
F5	0.5981		0.4443			0.316
F6	0.6234		0.227			0.158
F7	0.4347		0.3412			0.2246
F8	0.6221		0.508			0.4531
F9	0.5671		0.5345			0.4117
F10	0.5103		0.3716			0.3625
Average	**0.544**		**0.4100**			**0.3238**

Fig. 1. To record sound the sound wave using PCB 130 A24 Microphone
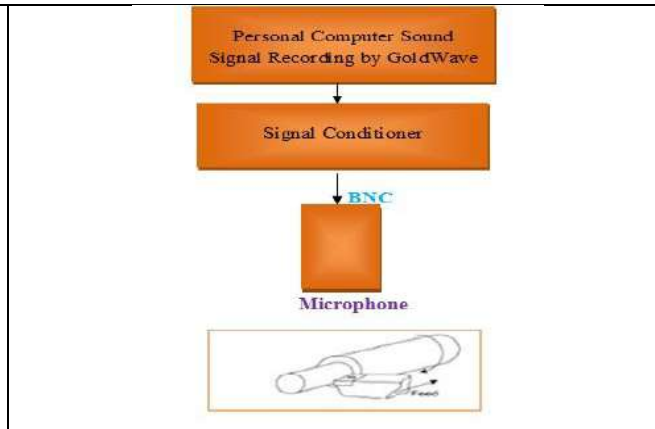
Fig. 2. Diagram of recording process during the machining process
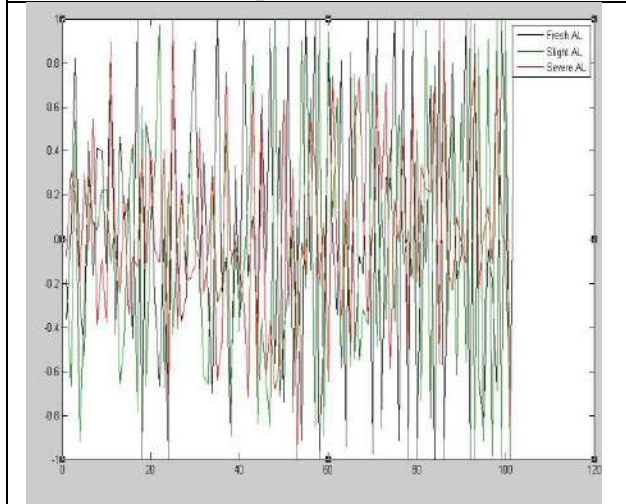
Fig. 3. Emitted Sound Signals of fresh, slight and severely worn conditions through Aluminium tool bit
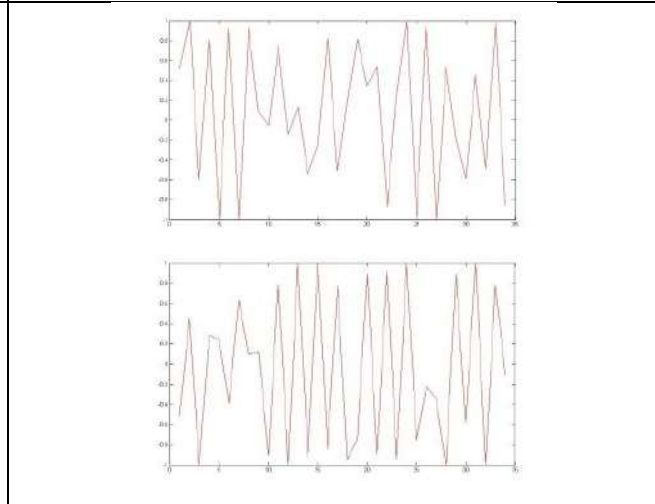
Fig .4.The frequency and wavelength analysis of fresh tool bit sound

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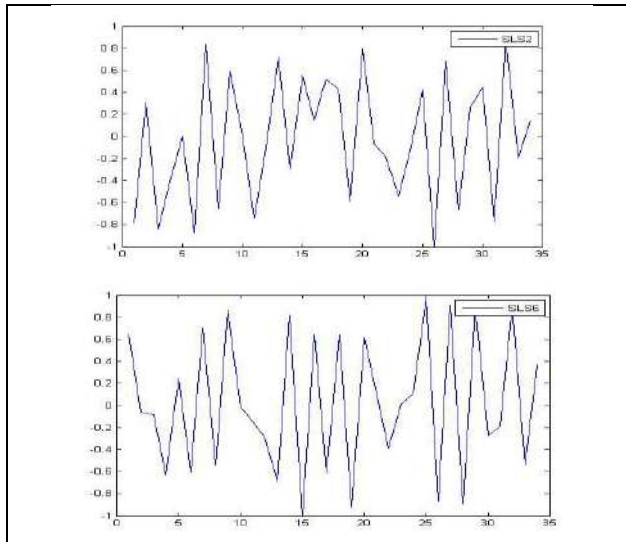
Fig.5.The wavelength and frequency analysis of slightly worn tool bit sound
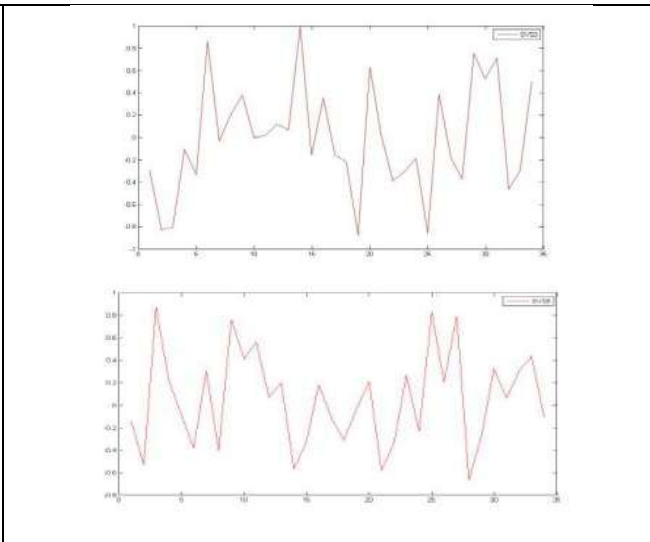
Fig .6.The wavelength and frequency analysis of severely worn tool bit sound
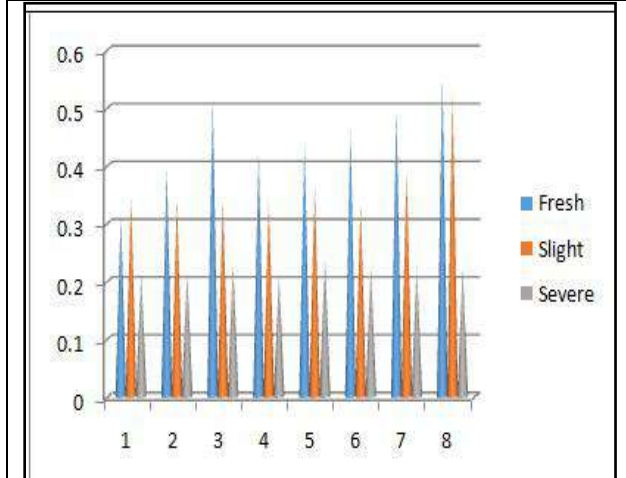
Fig.7. Averages of tool bit sound for fresh, slight and severe wear categories
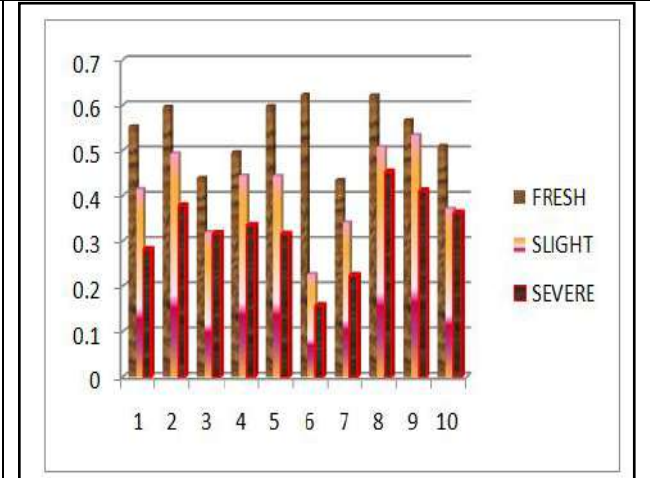
Figure .8. The significant difference between fresh and slight, slight and severe and fresh and severe wear categories

RESEARCH ARTICLE

Scrutiny of creativity and Artificial Intelligence in Real world

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ABSTRACT

Creativity is an act of turning new and creative thoughts into fact. Creativity is characterized by means of the potential to understand the sector in new issue. Artificial Intelligence (AI) is an extensive-ranging branch of laptop science concerned with constructing clever machines able to performing duties. AI approaches can generate new ideas in three ways: by generating possible combinations of previously known ideas; by generating novel combinations of previously known ideas; and by generating unique combinations of previously known concepts. AI is solitary of those buzzwords that get about all the time. AI or specialized programming software is an area of studies that seeks to make the pc smarter. They work on their very own encryption. It is the one in ubiquitous Jargon AI utilized in marketing, buying and fashion. It could also be cast-off to come across credit score card fraud, driving. AI seems logical that day or every different computer will rule round.

Keywords: Computational creativity, Artificial intelligence, Artificial intelligence creativity.

INTRODUCTION

Creativity in AI (artificial Intelligence) is a phenomenon in which something new and by some means valuable is shaped wherein as it's far the fundamental functions of human intelligence which is a assignment for AI. Computational creativity is the have a look at of building software which famous conduct that could be deemed creative in human. It's far used in autonomous creative assignment, which include driving motors, portray photos and so forth. Laptop chess was as soon as considered the "Drosophila of AI", nonetheless, fixing chess isn't currently viable by modern computers. It has motivated the manner wherein chess games are played on the top stage. Furthermost, the amazing grand grasp chess gamers use this contemporary AI chess engines to scrutinize their video games and the video games in their competition. This article explores the knowledge of creativity within the subject of synthetic Intelligence. IBM deep blue surprised the arena through creative Garry kasparov who is engaged into thought the nice magnificence chess player. The computer systems not simplest excelled human beings in chess or by portray, wager in lots of factors by using developing songs, writing articles Natural Creativity Natural Creativity is ready observing into the numerous theories at the posterior of creativity, examining studies, and exploring the present day machine getting to know techniques. You may test with creative

equipment that are powered by the contemporary system ahead understanding of techniques and behavior a complete iteration of layout wondering studies that may be applied in several exclusive business situations. You'll consciousness on what way the notion of inventiveness has advanced through the years and the way it has been examined. Then, you may actively have interaction and cooperate with others within the elegance via dialogue activates and have a possibility to pensively write approximately a scheme that evokes your creativity. For the duration of the direction, you may step far away from your laptop, plan, and behavior one generation of a layout assignment rub in the methods and models you simply found out, permitting you to benefit unfathomable visions on how humans are innovative in your personal neighborhood environment.

Creative Artifacts
Innovative Artifacts is ready escalating the description of inventiveness, from multi-disciplinary views along with computer technology, sensibility and creative practices, into fields like non-public creative practices, company improvements, and concrete rules. You'll have a look at famed performers who use the modern system mastering strategies in a persuasive manner to enlarge their innovative capacities. You'll additionally study unique business techniques and financial rules that goal to make groups and cities greater innovative. Formerly, keenly involve and cooperate with others in the session over discourse activates and partake a chance to pensively inscribe around a creative undertaking that conjures up you. Through this way, you will manage your personal layout research venture on the subject of your excellent, over more than one iterations that could interpret to tortious visions and enterprise strategies.AI enhance the inventiveness of human beings in art, track, dance, layout, process building, and distributing.

Art
AI is having an impression on the biosphere of visual art in a variety of ways. It has the capability to control current art, as it turned the Mona Lisa into a living portrait AI. It has built that resemble real people that may be found on the website, Harry Potter. This character no longer exists and even creates one-of-a-kind mechanism of painting.

Music
David Cope has worked for Trials in Musical Intelligence for the last 30 years. Manage a traditional musician and composer who resorted to computers in 1982 to help him break through composer's block once more. Since then, his algorithms have developed a amount of unique structures in a range of genres, Emily Howell as well, an AI that can make music entirely based on her own particular style rather than simply reflecting the styles of the previous day's composers. Dance and Choreography Using artificial brainpower as a collaborator was a strong approach for dance

choreographers to break out of their usual techniques. Wayne McGregor, an award-winning British choreographer and director, is known for use of technology in his work and is particularly interested in how AI can improve the dance in a mission with Google Arts Culture Lab. The usual of guidelines was built using thousands of hours of film videotape of dancers on behalf of both human styles. The AI then moved on to paintings, where it found a method to dance. The goal isn't always to substitute the choreographer, but to quickly iterate and expand different choreographic options.

AI AUGMENTED DESIGN

Industrial layout is another creative endeavor that AI has proven to be competent at. The initial chair designed using synthetic intelligence and put into manufacturing became offered at Milan layout Week, thanks to a collaboration between French fashion designer Philippe Starck, Kartell, and Autodesk, a three-D software programmer company. Any additional project that examines human-machine co-creation is involved in the Chair project.

Recipes

AIs innovation is also redefining the kitchen, not just by modifying long- standing recipes similarly by developing completely novel nutrition combinations in partnership with certain of the most well-known names in the food industry. Our favorite beverages might be given an AI makeover. Pre-orders for AI-evolved whiskey are currently available. Artificial intelligence is also presence used to learn about the brew masters choices. MITs Computer Science and Artificial Intelligence Laboratory (CSAIL) is using altogether the food photos we post on communal media. These food photographs are assessed with the practice of CPU visualization to people's eating habits and to recommend recipes using the food pictured. Write Novels and Articles Even if around is a ration of textual fabric to inform synthetic intelligence algorithms, writing takes been a difficult skill for AI to acquire. Even though AI has proven most successful in producing short-form formulaic content, such as journalism's & who, what, where, and when; testimonies, its powers are still expanding. AI has now written a unique, and despite the detail that neural networks constructed what many could consider a strange examine, it was nonetheless competent to doing with the news that an eastern AI software's short- form novel was close to winning a national literary contest, it's easy to get in what way it won't be long before AI can compete with humans in creating captivating content. Kopan's page has been updated. Superhuman Innovation is a book about artificial intelligence that stayed inscribed in collaboration with AI. Poem portraits is a group of AI and human collaborative projects.

• Through investigating the capability of conceptual domains
• By enabling previously unattainable eras through change

Modeling the stage of novel concepts can be a lot easier for AI than automating their evaluation. 1998 Elsevier Technology B.V is a [1] step closer to artificial intelligence (AI) creativity, which I define as the usage of autonomous computers to generate shockingly original, but appropriate, ideas, problem solutions, or other outputs. I argue that organizational researchers of creativity and innovation should devote significant resources to researching AI and computer-assisted human intelligence, the conducts in which they could produce innovative breakthroughs, and what way those advancements might affect and be influenced by people, clients, societies, and humanity. [2]

FOUR PERSPECTIVES OF CREATIVITY

• **Character:** Living organism agent who is thought to be innovative. Character philosophies investigate about the agent that inspires them to be creative.
• **Method:** The customary of inner and outer schedules engaged by the agent when creating an inventive artifact. Procedure theories aspect at the kind of actions that take place during the creation of creative artwork.
• **Product:** a thing, such as a painting or a mathematical theory that is realized as innovative or as the result of creativity.

Product theories investigate what it is about a product that qualifies it for the title of creative. The press is the encircling way of life that has an effect on people, strategies, and products, judging them as creative or uncreative. Press theories investigate what causes a person's lifestyle to regard something as new. [3]

TAXONOMY OF CREATIVITY IN AI

There are three main methods in which AI can be utilized to make novel notions:

• Making innovative combinations - making unique combinations is perhaps the finest of the three to be explored on a computer, and it's quite similar to making analogies or jokes.
• Investigating the capabilities of abstract areas is the things twitch to get exciting: imagine feeding an AI with musical ; grammar the fundamental rules of music composition, as glowing as a tilt of autographs from eminent composers like Beethoven and Stravinsky, including their mannerisms, regular harmonies, and melodies. This is what makes Beethoven's music sound like Beethoven's music. Then ask this AI to compose a new song that sounds like it was composed using Beethoven's music. David cope accomplished something similar with EMI (Experiments in Musical Intelligence).
• Making modifications - a project that dates back to the 1970s.

Novel Combination
 The first kind creativity is novel aggregate. When we consider about inventiveness to consider additional things, something from artwork or track to technological innovative innovation. One issue that all innovative acts have in commonplace is that they involve that novel aggregate that is the associations in our mind of current thoughts to form new ones. Innovation can immerge from novel combinations of or more existing ideas from low tech to excessive tech linguistic creativity, like every creativity, is set novel combination that is an idea that goes again at the slightest as some distance as the enlighten truth seeker David Hams. In his, enquiry concurring human information", published in 1748, linguistically, some novel mixtures are more gratifying. Who can resist a new punch-line to an antique funny story, which is closed to making jokes? "I don't consider glowers." They're continually as plentiful something this isn't so humorous, but a few common sense is over it. In 1996, AI PhD Kim invented jape, a computer programmer capable of making jokes. Although the jokes aren't amusing, we can see how a computer could make them. The second and third kinds are closely connected and comparable. Theyre "exploratory" and "transformational", creativity exploring the probable of conceptual areas is nothing but a geometric shape that represents an amount of first-rate dimensions. Relationship between the abstract and the literal. For example, conceive serving an AI with musical grammar, fundamental policies of song composition, and an extended list of signatures from famous composers, then teaching AI with simple track composition guidelines, and then asking AI to pen down. A new-fangled song that sounds exactly similar imagined. This is exactly what David 23cope achieved with EMI, and what makes this notion unique is based on pre-existing standards and signatures. Not only can the computer uncover other alternative mixtures, but it's also possible to mark it sound like a specific composition that hasn't been explored yet.

The third is undergoing change, but it has the greatest likely for disruption and will be further investigated in forthcoming years. It is the practice of integrating AI into all areas by employing virtual transformation via AI. An early instance of an automated hit was discovery device from the 1970s that would generate and redesign little portions of code. Synthetic development is the term for this kind of advancement. This has a large capacity, whereas computers today or in the upcoming are mostly attentive on the primary.

Computational creative
Computational creativeness is an area attempts to gain creative behaviors from pc, it is the connection of the fields of artificial Cleverness, cognitive phycology, philosophy, and the humanities. As mentioned computational creativity is observe of constructing software which well-known shows the performance of innovative in human. This kind of software program could be cast-off for innovative independent duties. Computational creativeness is a totally active

concern place, with many troubles still open to details. Building creative software provides each a technical challenge and social one. To continue in addition to embrace the point that computer systems aren't human. Creativity seems mysterious because whilst innovative thoughts is actual hard to bitch that how we were given them and we frequently tell the detail that it is not aware of how a creative concept manifests itself [3]. For in addition studying regarding computational creativity in genial, I endorse the AI mag unique problem on computational innovative.

Computational creativity in Music

AI has performed a crucial purpose in the records of pc music, nearly in view that it's starting within the Fifties. Most effort in Compositional and improvisational device and little effort has dedicated to expressive performance. The reviewed selection of an insufficient enormous fulfillments is in AI technique to music. The presentation of composition and song is an improvisation with a song performance and improvisation with an importance on the presentation of expressive music. For every tactics by AI still works which have proposed answers, describing what still desires to be achieved and a few feasible instructions for similarly studies.

Robot Creativity

There can be a growing form of information about robotics and inspiration. Robotics in lecture halls might be a potential novel tool for dealing with faculty creative crises, we frequently see a harm of theoretical advancement in the perception of creativeness and the devising mechanisms involved. First present an overview of prevailing studies on the usage of academic robotics to stimulate creativity in this post. We were show that in this part of effort, the precise mechanisms endorsed by robotics sports are rarely discussed. The effect of designing and coding robots on students innovative output, we employ a confluence version of creativity. In the framework of current trends in inventive cognition, the cognitive accompaniments of the procedure of building and software design robots. It deals with the issue of Meta intellectual and developing reasoning.

TECHNOLOGIES AVAILABLE IN AI CREATIVITY

Natural Language Generation
◎ Speech Reputation
◎ System Gaining Knowledge of Platforms
◎ Virtual Agents

Selection Management
◎ AI Optimized Hardware
◎ Deep Studying Structures
◎ Robotic Method Automation
◎ Text Analytics
◎ Bio Metrics
◎ Cyber Defense
◎ Content Material Introduction
◎ Emotion Reputation
◎ Photo Popularity
◎ Advertising Automation

SEVEN SIGNIFICANT OUTLETS OF SYNTHETIC INTELLIGENCE

Machine studying
Supervised gaining knowledge: In sort of studying, information specialists feed labeled training facts to algorithms and define variables to algorithms for accessing and locating correlations. Both the input and output of the algorithm are particularized/described.

Unsupervised learning: This form of mastering consist of algorithms that train on unlabeled information, an algorithm analyzes datasets to attract significant correlations or inferences. For instance, one approach is cluster evaluation that uses exploratory facts evaluation to attain hidden or grouping styles or companies in datasets.

Reinforcement studying: For coaching a laptop device to fulfil a multi-step method for which there are honestly defined rules, reinforcement studying is practiced. Right here, programmer's layout and set of rules to achieve a mission and give it fine and terrible signal to act as set of rules execute to finish the venture. From time to time, the algorithm even determines on its personal what motion to take to move beforehand.

Neural networks: Neural networks are considered to paintings much like the human brain does. In the circumstance of spotting handwriting or facial popularity, the brain very quickly makes a few selections. For instance, in the circumstance of facial recognition, the brain may begin with miles girl or male

Robotics: Robotics is an inter-disciplinary division of processer technological know-how and business. Robotics includes design, production, process, and usage of robots.

Expert systems: structure experts is a Pre Engineered constructing structures & Mini , Multi garage buildings, Structural steel design, detailing services issuer globally.

Fuzzy: A fuzzy set is a collection of objects with a series of club grades. A membership (characteristic) function is used to characterize such a hard and fast, which assigns a grade of membership to each item ranging from zero to one. Natural Language Processing Natural language processing (NLP) is an area of computer technology specifically, the artificial intelligence (AI) department.

METHODS FOR MUSIC GENERATION
Many procedures and tactics to adjust in number of technology [10]. However, categorized into a few main categories. We use the only subdivision used on previous analyses .Further current reviews expanded precise subsets of this taxonomy's six lessons. To enhance a seventh category for retailer-based structures is a meta-method that has gained a portion of traction and earns to be addressed one by one. The following are the seven classes:

Markov Chains
◎ Formal Grammars
◎ Rule/Constraint based systems
◎ Neural networks/Deep studying
◎ Evolutionary/Genetic algorithms
◎ Chaos / self-similarity
◎ Retailer's primarily based systems.

Each of those techniques is explained by referencing works that used mgs tactics that fit into those categories and a brief discussion of how those procedures might be perceived from a process perspective utilizing Boden's creative classes.

Industry Applications of Artificial Intelligence
◎ Civil Security
◎ Transportation
◎ Public Security
◎ Autopilot
◎ Smart Robot [11]

How artificial intelligence is Transforming the world AI is an actual broad discipline and nevertheless maximum of them aren't much acquainted with it. Whilst 1500 senior business leaders in US in 2017 were asked about AI, best 17% said they have been familiar with it. An amount of them were now not sure what it became how it might take a consequence on their unique organizations. They understood has been large ability for   alerting commercial enterprise method [12]. However were no longer clean how AI resolve the deployed within their personal organizations. AI is an era this is reworking every stroll of life. AI is wide variety of tool that allows humans to rethink how we combine statistics. AI is already changing the creation and rising important query for the society, governance.

CONCLUSION

AI approaches remained to make novel thoughts in three ways: by combining known concepts in unique ways, discovering the possible of conceptual spaces, and transforming ideas that were previously impossible to generate. Creativity is often characterized as a way for developing a novel and beneficial product, expression, or idea. Artificial intelligence's ultimate moonshot could be creativity. AI has already aided in the formation of pop ballads, imitated the techniques of famous painters, and inspired creative filmmaking decisions. Creativity can permit us to develop compelling paintings or progressive merchandise in an agency is another shape of human artifact similar to AI. Throughout those potential papers, evaluation the sector of creativity research from a cognitive and factor to the present development of AI model. Innovative concepts have been   generated by way of AI applications. The last suspicion of AI creativeness be able to a program that generate novel thoughts with is repealed as, however it become in a position to steer is indeed treasured.

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RESEARCH ARTICLE

Developing Scientific Temper on Health among Tribal Women in Tamilnadu

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ABSTRACT

Developing a scientifically tempered citizen on health is a challenge especially among the tribal population. The study is significant because tribal people have been identified with many health issues, and also attempts to identify existing levels of scientific Temper, awareness and to develop a temper on health science among tribal women. Intervention was conducted through presentation and interactive sessions to inculcate scientific health literacy. Data was collected by self-structured questionnaire including demographic data, their health practices and health issues, in-depth interviews were conducted. The findings of the study revealed that they were still using the traditional methods of treatments for minor problems. However, they have accepted that modern health services are important for their health issues.

Keywords: Scientific Temper, Health Literacy, Malayalis In Kolli Hills, Pachamalai And Kani Tribes In Pechiparai

INTRODUCTION

Tribal population has traditional knowledge on health and hygiene. This indigenous knowledge transferred from generations to generations through non-formal education by themselves, did not require any communication on health and nutrition from the outside world until recent decades. Access to modern media devices including TV and Mobile are present among them. Many studies have found that these technology devices had significant impact on their life. In India at least 70% of the females are engaged in farming activity including animal husbandry however,

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only less than 10% of land they own. Especially, tribal women share the burden of agricultural activities to with men (Shamna et al., 2018, Mohanta, 2017, Pillsbury et al., 2014). They work in the agricultural fields and related activities about 12 to 15 hours a day apart from household works. Intense workload at home and fields leads to various health risks to them (Badodiya et al., 2013). As Saikia (2013) explains, tribal health is a mystifying turf and a huge heterogeneity can be seen in tribal health, and in their convictions and practices. The tribes are living their life nearest to nature but it is affected more by socio-economic, socio-cultural and ecological measurements in their therapeutic practices. Their belief about sickness identifies with death and believes in sickness to the curse. Pacification of the god and annihilation of insidiousness spirits shapes a significant part of psychosomatic healing in their health knowledge (Mahapatra, 1994). According to the Article 51A (h) of Indian Constitution, it is the obligation "to develop scientific temper, humanism and spirit of enquiry" for each and every citizen of this country. The term "Scientific Temper" was introduced by Jawaharlal Nehru, first Prime Minister of India in his book "The Discovery of India". Scientific temper refers, "the adventurous and yet critical temper of science, the search for truth and new knowledge" (Nehru, 1946). Nehru understood that scientific temper is very much important in building the country's socio-economic development. During his period, the government had introduced various measures to develop scientific temper to get rid of myths and superstitions even on health conditions. This study was necessitated as tribal people have been identified with anemia, UT infections, diabetes and other diseases. Therefore, we should create awareness about such diseases which enables them to become more health conscious. The objectives of the study were to identify existing levels of scientific temper, increase of awareness and to develop a scientific temper among tribal women. In this study, it was hypothesized that tribal women have a significant knowledge on health.

REVIEW OF LITERATURE

In her study, Soma (2011) surveyed women in West Bengal about their socio-economic conditions, nutrition, health and impact of science education on their attitude to scientific temper. Results from the study revealed that science education had a significant improvement in modern society. However, their socio-economic status, health and nutrition status had a limited influence in respect to scientific temper. There are lot of work has to be done for to develop a scientific temper for better quality of life. Gigoo (2009) pointed out that tribal people are entirely different from other communities of social settings. The reason for most of their health care issues were from ignorance, poor facilities, and inadequate sanitation and furthermore, certain superstitious traditions and customs in their community. Usually in all tribal society, there is an abundance of fantasies identified with wellbeing. Health and healing are closely related to each other in their ecological environment. Tribes are utilizing various pieces of a plant, to cure of sicknesses, on the other hand they use it for populace control too (Chaudhuri, 1990).

The tribes offer significance to discover the reason for disease as opposed to the fix. In the wake of finding out the reason for disease, then only they go an treatment method. In their method, it is often seen that disease and treatment are closely linked with the magico-religious convictions (Mishra, 2002). According to Praharaj (2009), invasion and interruption by non-tribal factors were the major reason for fast diminution of normal encompassing and eco-system in tribal areas. However, the conventional medical system still discovers its significance of endurance in tribal area. They can still believe and relate to the conventional drugs, healers and practices of unnatural creatures. This is the reason for the existence of the uniqueness of tribal society. They feel comfortable with the security given by their customary healers against psycho-social issues and religious timidity. Meaning and significance of health is different for each and every individual. Generally people think that wellness is simply a nonappearance of illness. Since the development in health care industry, it is important to educate its citizens through any kind of media and make sure people progress in their health behavior (Sachdeva, et al., 2015).

METHODS

Using purposive sampling method, the present study was conducted in the tribal villages of Pechiparai Hills, Pachamalai Hills, and Kollimalai Hills. Most of the population depends on agriculture as their main source of income. Pechiparai is located in Kanyakumari district of the Western Ghats. KaniTribals are living scattered in and around 69 hamlets in the hill area. There are no proper transport facilities to reach the tribes. Most of the women from these tribal villages were working in rubber tree plantations, tapioca farms, banana farms and cashew farms. Pachamalai is located in the north western part of the Tiruchirappalli and Salem districts of the Eastern Ghats. The Malayali tribes spread over about 60 hamlets in the Pachamalai hills. The major crops grown are cassava, paddy, mango, castor, grapes, banana, silver oak and millets. kollihills is a part of Eastern Ghats and is located in the Namakkal District. There are over 300 hamlets in Kolli hills panchayat union. Most of them living in Kolli hills belong to the community of "the Malayali" tribes. Important crops such as cassava, paddy, jowar, bajra, millets, pulses, fruits and vegetables are cultivated in Kolli hills.

Scientific temper intervention was conducted through presentation and interactive sessions in three different locations on three different dates. The intervention classes were conducted by experts in women health by using demonstrations and pamphlets. Each session was two hours long with interactive sessions with participants. The intervention dates were 24th and 25th of July, 2017 at Kolli Hills, 26th and 27th of July, 2017 at Pachamalai Hills, and 30th and 31st of July, 2017 at Pechiparai Hills. The study subjects were 66 tribal women with the age group between 18-65 years were selected. Data was collected by self-structured which includes demographic data, their health practices and health issues. In addition, in-depth interviews were also conducted to understand scientific temper and their traditional health practices after the intervention session. The sample size for in-depth interview was 6, two each from villages of Pechiparai Hills, Pachamalai Hills, and Kollimalai Hills. The purposive sampling was used for the interview. Frequency and percentage analysis were done using SPSS. Chi square test was utilized for correlation and p values were determined.

RESULTS AND ANALYSIS

Main reasons for not accepting the facilities, especially health related government initiatives, were because of myths and misconception in their communities. Therefore, to teach them the importance of health and hygiene, the intervention sessions were conducted for women in above mentioned tribal areas. Most of the populations in tribal societies are illiterate and deprived, they lack modern health facilities. The lack of knowledge about the importance of vitamins and calcium, and poor eating particularly in pregnancy also reported. However, their medical practices cannot be defined or understood only in terms of scientific aspect, because they are largely influenced by their social and cultural perspectives. They have a very rich social and cultural heritage even though they live in poor conditions. Table 1 reveals the demographic characteristics of respondents. A total of 66 participants from three locations were involved in this study. According to the age group of the respondent, 37.9 per cent of women were below 30 years, 45.5% of women were aged between 31 and 50 years, and 16.6% of them were above 51 years old and 83.3 per cent of them were married in this group of participants. Around 30 per cent of participants were illiterates and the remaining 70 per cent of participants had formal education. In this group, 93.9 percent of them had access to health care facilities in their place.

The above table 1 shows participants' households behavior and facilities related to health. It shows that participants had good hygiene behavior. According to the table, only 65.2% of participants had a bathroom facility at home. The remaining 34.2% of participants were using open area for bathing. And only 62.1% of household had toilet facility at their home. Again, the remaining 37.9% of participants were defecating open. And, there was no good drainage facility in their household area. Ninety seven percent of participants were living with no drainage facility. According to table 3, the results revealed that 57.6 per cent of participating tribal women were using sanitary pads as absorbent. Whereas remaining 42.4 per cent of women prefer clothes and some other combinations during menstruation. In

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account of disposal of absorbent, 69.7% of women were using incinerating the material. Another 24.2% were burying and 6.1% were using public dustbins for disposal. Large samples of participants were treating themselves when needed, however 34.8% of women seek help from their mother/mother-in-law and 10.6% prefer to get help from a gynecologist.

In the table 4, taking pills for body pain is common among this group. Just over 62% of participants were taking pain-killer medicine with doctor prescription. However, without a doctor's prescription a lot of them use pain-killer medicines. The reasons might be because they work in both household chores and farmlands. Mostly they had used it to reduce pain caused by over workload both at home and farm. According to Table 5, the results of Chi square test shows that there was a significant relationship between television watching and health literacy (p < .05). Thus, the proposed hypothesis was accepted. In fact, there was strong statistically significant relationship shown which explains that watching television has contributed to participants' knowledge about key health issues they have encountered. However, there were no considerable impacts of the other two mediums; newspaper and radio in order to increase participants' knowledge about health issues.

In-depth Interviews

The aim of any scientific temper campaign is to identify the target that might be a single, families and the community itself. Target is necessary because to achieve the objective of any intervention program health-program needs a clear planning and implementing intervention program to modify the behavior that needs to be changed. The behavior change and adapting to new ideas will not occur immediately. There are several processes that need to finish on various stages. After the classroom session by the experts about health and hygiene, the following Were an excerpt from the in-depth interview. According to Maheshwari (Personal Communication, July 25, 2017): "*Trainer explained why science is important and how to gather knowledge about science. They have also explained what health is and how and why it should be taken care of, especially for women. She further clarified our doubts about medical issues such as anemia, breast cancer, and vitamin deficiencies. She advised us not to ignore even the smallest problem related to health. We have learnt that the basic understanding of science and its importance in our health practices*"

A 36 years old female from Pachamalai Hills, Sudha(Personal Communication, July 27, 2017) mother of two explained: "*The classes were good and important for us. Nowadays, diseases are not cured with traditional medical practices. We have to travel to a nearby primary health care centre. Or we have to wait till a doctor to us once in a while. Instructor taught us how to manage ourselves. They taught us few daily practices to follow for better hygiene. The training in health enables us to identify and handle the common illnesses within home itself. Earlier we believed that diseases like smallpox, plague etc. were associated with supernatural power. We also believed that epidemic diseases were caused by the annoyance or rage of god*". A 20 year's old unmarried girl from Pechiparai Hills, Preeja Mol(Personal Communication, July 31, 2017) explains that:"*This training was real eye opening for all women in our community. We have very low level of knowledge about menstruation and UTI. We cannot openly discuss about it with parents so we don't know the correct information about menstrual and personal hygiene. During the training, they have explained about menstrual hygiene and practices to follow for healthy practices*".

A 56 years old woman residing in Kolli Hills Rajeshwari (Personal Communication, July 25, 2017) reveals that:"*We believed in supernatural causes for the incidence of all illnesses. A warm relationship with the divinities and familial spirits can guarantee good health for us. If we break the rules, they will get angry and send sicknesses and different catastrophes towards us. We believed that there was a link between sickness and spirits. The evil spirits were reason for most of diseases and if we perform pujas, they can go away and automatically diseases get cured. But now we understood that we should not consider superstitious beliefs during life threatening conditions. A 60 years old female from Pechiparai Hills, Lakshmi Personal Communication, July 31, 2017) describes their beliefs: "We have profound trust in the efficacy of our doctor (not a qualified doctor). We first contact him and he can identify the reason of the disease by prediction. He used to treat us according to our customs. He also uses herbs to treat sickness which is very helpful. Earlier we used to believe that our traditional medicine can cure all diseases. In addition, it is economical and readily available. We also believed that there was no useful treatment for a few*

diseases like chickenpox and thought that those were created by supernatural powers. But now we realize the importance of health and hygiene".

Another participant, a 25 year's old educated girl from Pachamalai Hills, Saranya (Personal Communication, July 27, 2017)expresses that:*"We believed that black magic, witchcraft and sorcery can cure illnesses and protect from evil spirits. Even though we still believe in supernatural powers, modern medicine is very much essential for us now. We follow maternity and child health practices now including immunization. We can get free treatment for all types of illnesses at the government hospitals. We used to have no knowledge about personal hygiene, but now we pursue that also. We have some good traditional herbs to treat specific problems and we have modern medicine also. I think both can coexist in our livelihood".*

Attention to tribals health has not been sufficient enough. Because generally people thought that they live very close to Mother Nature which is helpful for good health. Another reason could be that few tribal are not ready to accept modern health practices since they still believe in supernatural causes. Even though modern medicine is alien to them, it has a massive impact on tribal society. They have their own insecurities for this new system and they considered that it was not meant for their customs. However, education such as training programmes influenced them to accept to use of modern health care. It might be consistent the statement that tribal health beliefs are made out blindly, yet sometimes confidence and healing might be a piece of the treatment (Singh 1995). Tribal medicine is to a great extent old school and customary which is passed on to one member of the family to another. The tribals have a good learning of basic illnesses and its therapeutic reaction to medicinal plants and the roots of plants. They treat patients experiencing basic health issues such as fever, body pain, stomach issues, common cold and diarrhea. Then again, they believed illnesses like small pox, snakebite are caused by bad spirits and manipulation of evil spirits and present day medicinal treatments are viewed as useless. However, they have understood that both traditional and modern medical systems are increasingly useful. Along these lines, in the current conditions both ethno and present day medical practice assume an important part in the health care of tribals. Results of in-depth interviews reveal that scientific transfer intervention was effective which improved the level of knowledge about health and hygiene practices among the participants. The in-depth interviews also focuses on their health seeking behavior.

CONCLUSION

The investigation is an effort to understand different aspects of tribal health and hygiene with regards to Kani and Malayali tribes from Pechiparai Hills, Pachamalai Hills, and Kollimalai Hills. Hygiene is a significant part of good health which is identified with the vast majority of the parts of living. Generally, hygiene is considered as a safeguard measure to decrease the frequency and spread of illnesses. According to the results of this study, there were still few of them using the old methods of absorbent during menstruation. Prevalence of using pain-killers was common in this group. They had very good knowledge about common illnesses such as anemia, UTI, dengue, fatigue and prolapsed uterus. Their traditional medical practices were interrelated with their spiritual viewpoints. Even though traditional health practices and superstitions, especially local beliefs and customs hindered them to get a good health care system, they have accepted that modern health care was found to be increasing. The intervention also has shown their interests for increasing their temper on science.

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Table 1 Demographic characteristic of participants

Age (%)		Education (%)		Marital Status (%)		Health Care Facility (%)	
18-30	*37.9*	Illiterate	*30.3*	Married	*83.3*	Yes	*93.9*
31-50	*45.5*	Primary	*37.9*	Unmarried	*16.7*	No	*6.1*
51 & above	*16.6*	Matriculation	*21.2*				
		UG/PG	*10.6*				

Table 2 Household hygiene of participants

Medical Condition	Frequency	Percentage (%)
Bathroom at home		
Yes	43	65.2
No	23	34.2
Toilet at home		
Yes	41	62.1
No	25	37.9
Drainage		
Nil	64	97
Open	2	3

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Table 3 Menstrual health behavior of participants

	Frequency	*%*
Absorbent Used		
Sanitary Pad	38	57.6
New Cloth	1	1.5
Old Cloth	15	22.7
Some Combination	12	18.2
Disposal of Absorbent		
Burying	16	24.2
Public Dustbins	4	6.1
Incinerating	46	69.7
Seeking Help		
Self-Treatment	36	54.6
Mother/MIL/Relatives	23	34.8
Gynecologist	7	10.6

Table 4 Taking Medication

	Frequency	%
Taking pain-killers with prescription		
Yes	41	62.1
No	25	37.9
Taking pain-killers without prescription		
Yes	47	71.2
No	19	28.8

Table 5: Knowledge of medical conditions with media exposure

Characteristics	Knowledge of Anemia	Knowledge of UTI	Knowledge of Dengue	Knowledge of Chronic Fatigue	Knowledge of Prolapsed Uterus
Watching TV	$\chi^2= 8.38$	$\chi^2= 9.64$	$\chi^2= 14.04$	$\chi^2= 17.63$	$\chi^2= 15.06$
	p= 0.04*	p= 0.02*	p= 0.00*	p= 0.00*	p= 0.00*
Reading Newspaper	$\chi^2=1.97$	$\chi^2= 3.09$	$\chi^2= 0.35$	$\chi^2= 0.69$	$\chi^2=4.45$
	p= 0.37	p=0.21	p= 0.84	p= 0.71	p= 0.11
Listening Radio	$\chi^2= 1.19$	$\chi^2= 4.98$	$\chi^2= 1.33$	$\chi^2= 7.19$	$\chi^2= 5.63$
	p= 0.75	p= 0.17	p= 0.72	p= 0.07	p= 0.13

* = p < .05

RESEARCH ARTICLE

A Study on India's Travel and Tourism Industry to Retaliate the Impact of Covid-19

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ABSTRACT

COVID-19 has played a major role in human economic behaviour, which has had a profound impact on the tourism industry and even globally. Immediate control measures, such as the necessary travel restrictions, to avoid physical contact, distance with the public destroys the interest in going to a different normal residential area and seeking access to tourism resources. At first people were afraid of this deadly disease and even got it. not realizing that travel can be a factor in its spread. Many people have started running their homes with the virus in those cities that were far from dangerous and COVID-19 has had an immediate impact on the entire economic and social sector and affected them. However, an initial approach is required to assess the current state of the situation and it is financially affected. It will go a long way in building an effective tourism industry epidemic management system.

Keywords: COVID-19; Economy; Pandemic; Tourism industry.

INTRODUCTION

COVID-19 has played a major role in human economic behaviour, which has had a profound impact on the tourism industry and even globally. Immediate control measures, such as the required travel limits, to avoid physical contact, distance and community undermine the desire to go to a different normal residential area and to seek access to tourism resources. At first people were afraid of this deadly disease and even got it. unaware that travel can be a factor in its spread. Many people have started running their homes with the virus in those cities that were far from dangerous and COVID-19 has had an immediate impact on the entire economic and social sector and affected them. However, the first method is required to assess the current state of the situation and is financially affected. It will go a long way in building an effective tourism industry epidemic management system.

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Tourism And Covid-19 According to UNWTO 2020 estimates, immigrants have dropped by 22% in the first quarter of 2020 and are expected to register between 60% and 80% annual growth, with losses of between US $ 910 billion and 1.2 trillion. In addition, social media has put pressure on the hospitality industry in which many resorts are forced to stop immediately. Among other types of businesses, tourism and its facilities are the main victims of the outbreak. With the outbreak of covid-19 in India, it had initially spread to all parts of the country. Because of fear of this deadly disease, many people did not realize that wandering could be a major factor in the spread of this deadly disease. More and more people are rushing to their cities with those deadly viruses and the main reason for the spread of covid. For the next 6 months they travelled in fear of covid-19 and the lockdown was introduced by the government across the country and was closely followed by the public to protect themselves from this deadly virus. This has led to a decline in foreign tourists and foreign trade hubs, which often attract with the help of a free trade agreement.

Here are some ways in which the government can revitalize the tourism industry:

1. The first step the government has taken is to have a share of open space for tourists.

2. The Government and the Central Government limit the number of guest gatherings from time to time in accordance with all covid-19 rules.

3. The Covid-19 test report was an important document that must accompany each visitor in order to travel.

4. The Covid-19 test should be taken 72 hours before the trip.

5. Currently, policy research has played a major role in revitalizing the tourism industry.

REVIEW OF LITERATURE

Tazim Jamal and Christine Budke: In this paper the researcher highlighted about global tourism affected by the epidemic: domestic and international responsibility and predictable action, based on the impact of MERS and SARS, which raises the need for more effective communication and revitalizing post-epidemic. . DR. HarshitDwivedi, DR. Ganesh Singh Rathore: The paper is heavily analysed by the impact of COVID-19 on various aspects of tourism - inbound, outbound, joint, MICE (Meetings, Incentives, Conferences and Exhibitions), travel and leisure - will continue to do poorly. until the next two hours. Also, with regard to "FAITH" which has been urging the government to achieve a goal-based self-confidence, a targeted tourism sponsorship fund is available so that tourism athletes can meet their operating costs and maintain jobs. DR. Amlan Gosh: This paper highlights the impact of the global tourism epidemic in India and focuses on strategies developed to address the problem by the Indian Tourism Area. This paper highlights the role of stakeholders and proposes a Regional-based tourism policy to improve the current crisis. Sandhya H: This study aims to understand the challenges faced by various players in the tourism industry in order to survive this epidemic. The study also sheds light on future prospects in the post-Covid context and some of the strategies taken by industry players to manage future demand in a more sustainable and secure manner. This paper makes sense and is based entirely on the literature review of various research papers focusing on the global Covid epidemic.

Objective

 • The main objective of this paper is to highlight that the travel and tourism in India is worstly affected by the covid-19 epidemic and how it will retaliate in the near future. India is considered as tourist hotspot and is attracted by many foreigners but due to the epidemic this situation has been completely disrupted.

• The other main highlight of this paper is to show that the tourism industry will retaliate in the near future with the help of the central government and government because statistically the tourism industry is considered the backbone of the Indian economy. It will largely will contribute to GDP growth

About The Study

 Tourism, would account about 5.06 percent of India's GDP (2016-17). Ongoing epidemics, travel restrictions, and nationwide closures have halted the entire tourism industry, and unlike other sectors, tourism will take longer way to recover, especially recreational tourism. This will have a direct impact on provinces such as Uttarakhand,

Rajasthan, Kerala, Himachal Pradesh, Goa, Sikkim and other north-eastern provinces that completely depend on tourism as a source of state revenue. During the Covid-19 violence, many industries were forced to adapt many new ways of doing business, as workers began to work from home and customers were told about online forums. While some industries are adapting with this change easily. Tourism and agriculture, was not able to make this change. Service-driven tourism and hospitality are an unparalleled industry that can sustain economic activity remotely. Employees and customers do not have access to these services remotely. One way in which this is reflected is the decline in the number of foreign tourists arriving in India from 12 lakh in December 2019 to 470 in April 2020. The graph presented below will clearly show the decline of the tourism industry. (According to research and records published this graph shows the collapse of the tourism industry by 2020 due to the covid-19 epidemic)

The Indian tourism industry employs about 8.75 crore people (12.75 per cent of the total population employed in 2018-19), such as people from the tourism industry, tourists, tourism workers, homeowners, drivers, directors, small traders, professionals and craftsmen. among a host of other service providers. The sector also has strong forward and backward connections to other sectors such as agriculture, transport, handloom, and FMCG to name a few. Disruption in the tourism industry will cause many people to lose their jobs. The food and hospitality sector are already struggling under the pressure of fixed costs and no lower costs. FAITH, a consortium of corporations of tourism organizations and the tourism industry has estimated the loss of Rs 10 lakh crore in the industry due to COVID-19. This will also have an impact on the influx of foreign tourists, which means a significant reduction in earnings close to Rs 2,10, 981 crores in Q1-Q3 2019. India will still face concerns in the tourism industry. (As per the research and records published this is the graph clearly shows the unemployment that is been faced by people in tourism industry during 2020 due to covid -19 pandemic)

Limitations
- The present study is been limited to only few aspects because it is impossible to consider each and every factors.
- The study is based on issued that is been faced presently by the tourism sector and the sources are considered to be valid for the research.
- The study assumes that the information collected are reliable and unbiased.

Suggestion
Tourism is one of the areas most affected by the epidemic. As planes landed around the world, countries were closed, people began to work from home. Most affected are air travel and international travel. The world's leading tourist destinations began to decline as international borders closed. This is the current state of tourism as the epidemic has affected us all and may be approaching in two years. Now the world is struggling to prepare for mass testing and vaccination. The concept of natural bubbles, social isolation, door closure, etc., all work with great flexibility in keeping the epidemic low. Soon life will be normal and the mask will be removed from the face.
How Can the Tourism Industry Improve Regeneration Strategies?

Revitalize Local Tourism: Enhance the power of local tourism by adding more local tourist attractions to tourist attractions. Adding more tourism destinations will help create jobs, new markets and business prospects in the immediate area. The development of local tourism is essential to redress the losses incurred by the government, tour operators, resorts and the entire tourism system. recreational activities that can handle a large crowd with appropriate covid-19 contracts. Villages and tourist attractions with extensive lounge areas, highways, remote restaurants, etc., will help to spread the crowd.

International Travel bubble: Creating travel bubbles is very important and already many countries including India have already introduced air bubble systems in neighbouring countries. Air bubbles are developed in line with the covid-19 protocol to reduce the global epidemic. Under the Air bubble program people who are fully vaccinated and perform Rapid PCR (Polymerase Chain Reaction) within 48 or 72 hours can travel the country. Therefore, the

International Travel Bubble will help revitalize the international tourism industry as people are able to travel to destinations safely and enjoy their holidays during the epidemic.

Vaccination Passports: In order to improve vaccination standards and to spread the value of the vaccine, many countries have enacted vaccination law to travel abroad, reside in resorts, and be out in public places. There are currently world-famous vaccines available to many people worldwide. To name a few are Pfizer, Moderna, Astra Zeneca, Covid Shield, and Sputnik. All are being developed by various medical research institutes around the world to curb the spread of the epidemic. During the trip, the vaccination passport needs to be shown to the various places where it is needed. It will facilitate safe travel in all countries and will serve as a step towards the development of the tourism industry.

New Special Offers: For people who like to travel, smart marketing plays a big role. Create public awareness about ways to control outbreaks. At the same time, encourage them to be given special provisions for parks, hotel stays, extra holidays, hotel work, and more. The sense of security at the same time had a measure of entertainment. In the tourism industry that is the case. It is important to introduce special packages so that they recover faster from their decline. So, in short, for the tourism industry to recover from a recession, they must reorganize the normal marketing and operational approach and follow the new tourism laws during the pandemic era.

CONCLUSION

India's tourism industry is growing and has great potential to create jobs and make huge foreign investment without providing additional information about the country's economic and social development. Eco-tourism needs to be promoted so that tourism in India can help preserve and sustain India's natural and cultural diversity. India's tourism should be developed in such a way that it welcomes and entertains visitors in a way that does not disturb the environment or harm the environment and preserves and supports traditional culture in the areas in which it operates. In addition, as tourism is a multi-sectoral service, and especially the services industry, it would be necessary for all central and provincial government agencies, private companies and voluntary organizations to be active partners in the pursuit of continued tourism growth if India becomes a global player. tourism industry.

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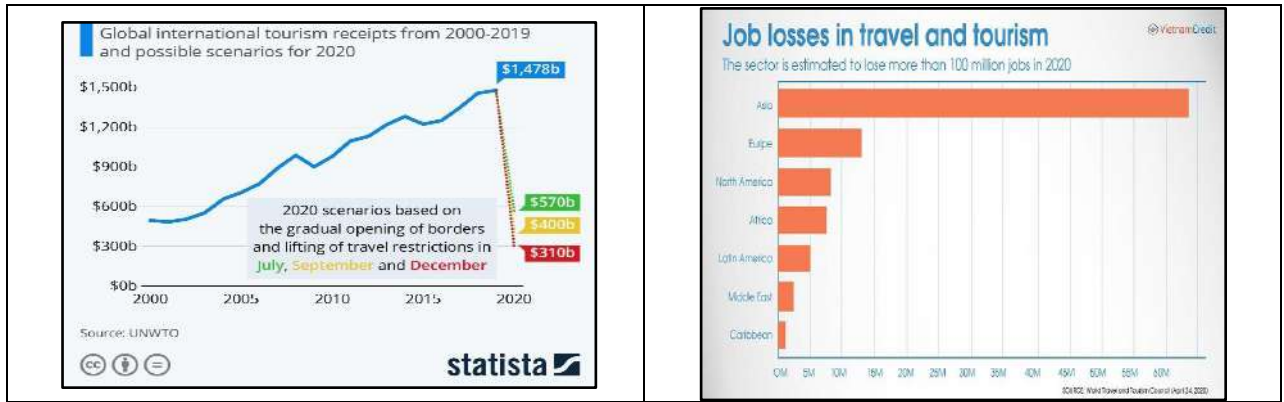
| Fig.1.According to research and records published this graph shows the collapse of the tourism industry by 2020 due to the covid-19 epidemic | Fig.2.As per the research and records published this is the graph clearly shows the unemployment that is been faced by people in tourism industry during 2020 due to covid -19 pandemic |

RESEARCH ARTICLE

Effect of Functional Postural Training in an Adolescent Spastic Cerebral Palsy

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ABSTRACT

So much focus is being given to impairment based physiotherapy intervention for cerebral palsy by the contemporary physiotherapist. When theoretically based interventions like Bobath's NDT emphasise tone based interventions, literature-based interventions always support functional based rehabilitation. We have treated a patient on a functional basis and presented in this report. The purpose of this study is to evaluate the effectiveness of exercise interventions that may improve postural control in an adolescent with cerebral palsy (CP). The patient has performed a functional evaluation with pre and post-treatment with the follow-up of about 3 months. We found that the patient with adolescent cerebral palsy was found to be improving significantly when functional postural training was provided and it enhance the lower limb ROM, muscle power & strength, coordination and also functional performance of their daily living.

Keywords: cerebral palsy, motor relearning, gait rehabilitation, muscle strength, functional performance.

INTRODUCTION

Cerebral palsy (CP)—is the most common chronic non progressive and infantile encephalopathy that affects the development and results in disabilities due to abnormal motor control1). Structural and mechanical changes in body alignment, as well as musculoskeletal changes, are developed therefore compensatory mechanisms are overcome by gravity and recruit new muscle groups to maintain stability. This lead to muscle imbalance, increased hypertonia, and deformities which affect postural balance and develop complex motor skills and impacts functional activities, thus limiting participation in different activities(2). Development of automatic postural reactions, including

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straightening, balance, and protection and also be delayed or undeveloped(3). Independent walking plays an important role in activities of daily living, improves bone density and cardiopulmonary endurance, reduces obesity, etc. Therefore, it is an important goal of clinical and community-based rehabilitation for many children with cerebral palsy(1). Children with spastic CP present with muscle weakness, spasticity, and limited selective voluntary control, resulting in gait and balance impairments(4). These gait abnormalities reduce gait speed and affect daily activities, sports and leisure, quality of life, and social interactions(5). Although most children with Unilateral spastic CP can walk without assistance, motor skill impairments should be addressed to improve gross motor function in everyday life. The LE intensive functional training (LIFT) protocol - integrated the principles of motor learning, such as intensive and structured practice, active participation, and skill/exercise progression.

CASE REPORT

A 16year-old boy was diagnosed with spastic diplegia cerebral palsy who came to the Rehabilitation department. The boy was 130 cm in height and 27 kg in body weight. The child was able to ambulate by himself with an assistive device (anterior rolling walker) and ankle-foot orthoses. On examination, we observed muscle weakness (Kendal scale) and spasticity (Ashworth scale) predominantly in lower limb and trunk; decreased range of motion and deficits in postural control, especially in sitting posture and impairments in functional performance, especially decreased gait velocity, poor alignment during functional tasks, difficulties in sitting unsupported and to perform lateral walk. Now the patient is assigned for physiotherapy treatment to enhance their muscle power, range of motion, coordination to improve functional mobility.

CLINICAL DESCRIPTION

Initially, the patient was assessed by a physiotherapist Once before intervention i.e pre-test and after intervention, post-test and a follow up session at end of 3-months. Then, brief information about the Physiotherapy given to the child's parents/guardians were briefed and the consent form was signed. The primary outcome measure was the 1-minute walk test (2MWT). This measure was selected based on the age of participant which causes decreased attention to task and ability to comprehend pacing required for 6 minutes. Secondary outcome measures were the 10-Meter Walk Test (self-selected and fast gait speed), 30-s chair rise22 (strength), single-leg stance on both the side(balance), and range of motion of all lower limb-joint has taken with a universal goniometer. The tone of all lower limb muscles were checked by Modified Ashworth Scale, Gross Motor Function Measure (GMFM-88) and Timed Up& Go (TUG) evaluation were conducted. GMFM is a valid, reliable and sensitive test, specially developed for children with CP. Here GMFM and GMFS-88 were also evaluated. It measures gross motor function improvements over time or as a result of the intervention. TUG is a practical and quick test that has been widely used to verify ambulatory mobility.

CLINICAL FINDINGS

Reduced ROM in the bilateral lower limb, muscle power, impaired functional mobility.

THERAPEUTIC INTERVENTION

At the first session(0-4 weeks), General range of motion exercises are taught without resistance. Once the exercise technique was mastered, the training load was determined. The training load was adjusted by adding free weights around body segments. Stretching also enhance to prevent joint stiffness and promote flexibility in each joint. The child was evaluated every week to adjust the training load. All exercises were performed in three sets of ten repetitions, with a 90-second interval between sets. For4-8weeks: Resistance training was started with the low-resistance bands. (yellow, red, green) As the strength is increased in the child resistance band colour was also changed with a high resistance band. The strengthening exercises were given to the lower limb muscles like hip flexors-extensors, abductors adductors, knee flexors-extensors and ankle dorsiflexors plantar flexors. 2MWT is enhanced to improve the functional capacity of the individual. The strengthening domain primarily involved LE exercises using body weight as resistance that targeted functional activities (example sit to stand, step-ups, vertical jumping, stair climbing). They performed sit-to-stand movement with support and walked with assistive devices (anterior roller walker). Time in seconds was recorded from the ''go'' cue to when the child sat down on the bench

was encouraged to improve their dynamic control.For8-12 weeks: Skill progression was used to make LIFT challenging and intensive enough to elicit changes in motor skills and function. Exercises targeting specific muscle groups important for gait were also included to a lesser extent with muscle-specific exercises (eg, bridges, clamshells, heel raises). The balance activities such as tandem walking, one-leg standing, and balancing on unstable surfaces and balance beams. The coordination domain involved activities such as ball kicking, jumping jacks, hopping through squares (hopscotch), and galloping/skipping. Gross motor function was tested with GMFM domains like standing, walking, running, and jumping was evaluated before and after sessions.

FOLLOW UP & OUTCOME

Initially from day 1 we started with normal range of motion exercises in bilateral lower limb along with this stretching is enhanced particularly for calf muscles hamstring adductor or butterfly stretch. Each exercise is performed with 3 sets of x10 reps. Each set of exercises with an interval of 90sec And the above program was followed for 4 weeks. From 4th week onwards we encouraged resistance training, targeted exercises to the bilateral lower limb. This resistance training is given to hip flexors - extensors, hip abductor - adductor, knee flexor - extensor, dorsiflexors and plantar flexors using theraband and weight cuffs were used. Targeted exercises mainly encouraged to improve functional performance and also to improve the muscle strength of the individual. Here targeted exercises like sit to stand, single-leg stance, leg presses, vertical jump, stair climbing and this exercise were also performed with 3 sets x 10 repetitions. Alone with this 2 Minute walk test enhanced to determine the functional capacity of the individual and was calculated by using a stopwatch, paper and pen to enrol the distance and time taken to complete the cycle. For each cycle of 2-minute walk test pre & post heart rate value, distance and time taken to complete the cycle were compared and contrasted to determine the efficacy or effectiveness of 2 MWT in CP. Initially, GMFCS(level 3) was noted which show moderate categories with the GMFS -88 scoring. The above programs was followed up till 8 weeks and the scoring were noted to determine the efficacy of the Functional performance in that individual. After 8 weeks we started with balance and coordination exercises, Specific targeted muscle group performance to enhance the gait pattern. Therefore, single-leg stance, tandem walking, leg presses, Hopping with different shapes were encouraged to promote balance and to improve agility. On comparing the GMFS -88 with the above session it has been improved and also the GMFCS level changes (level 2) based on the improvement with the above rehabilitation protocol, so we concluded that the functional postural training had a benefit on adolescent cerebral palsy.

DISCUSSION

In this study, functional postural training in adolescent cerebral palsy has shown a positive effect of structured exercises protocol about 3 months follow-up. There was an improvement in increase ROM in the bilateral lower limb, muscle strength, balance, functional performance. Distance ambulated during a 1MWT represents a potentially cost-effective and user-friendly method of assessing gait capacity in a clinical setting. This was corroborated by an increase in fast gait speed at both time points that approached significance in CP.30seconds chair rise test revealed a larger increase in functional strength in terms of lower limb functional training. Task-oriented practice. It showed greater improvements in activities performance since it enabled the refinement of neural control commands besides increases in strength(6). There were also improvements in GMFM scores, with major increases in dimensions D and E, which represent ''standing'' and ''walking, running and jumping'' items. These items are related to the exercises proposed during the intervention, such as sit-to-stand movement and stair-climbing.TUG test requires sophisticated postural control and dynamic balance since it requires a process of planning, initiation and execution of complex activities such as rising from a chair, walking, changing direction and sitting. Increases in muscle strength are accompanied by decreases in TUG time. If the decrease in TUG time means that child improved in agility and postural control since that minor time in the TUG test is associated with low risk and high gait speed [7]. Task-oriented weight-bearing (closed chain) strength training for children with cerebral palsy can be effective in improving functional performance.

PATIENT PERSPECTIVE

Initially I was not able to fulfil my daily routine even though I walked with an assistive device, I needed some external support to do them. After taking the functional type of physiotherapy treatment, I see a lot of tasks performed with less energy expenditure.

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RESEARCH ARTICLE

An Approach to Reduce Overloading in Distributed Clustering using VACC Approach

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ABSTRACT

Distributed clustering has a wide range of applications and its features are increasing day by day. However the most concentrated issue is load balancing and more number of researches is processed to overcome this issue. This paper proposes an approach to obtain optimal result from such a huge database (big data), and the way of handling load balancing with these combination it obtains better performance. Virtualized Ant Colony Clustering approach (VACC) is proposed in this system where the load is balanced through virtualization and task will be scheduled and processed. Work is balanced as well as overhead issue is also handles and retrieval of content on servers is obtained by soft computing techniques. It not only retrieve the content exactly it also obtains accuracy and efficiency in obtained results. The result obtained from this approach achieves better result when compared to existing available methods.

Keywords: Distributed clustering, Load Balancing, Soft Computing, WRR and Clustering.

INTRODUCTION

Distributed clustering is an enhancing approach due to its wide range of applications all over the world. Where a query is processed by the servers and combined together to produce effective results to users. One of examples to this is potential that lots of the services you run in your network today are portion of a distributed systems cluster. Domain Naming System (DNS), Windows Internet Naming Service (WINS), and Active Directory are all examples of

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services that are distributed; yet provide a service to users. In distributed clustering the objects are assumed to be resides on different sites. Without transmitting all the objects to a federal location (represented as server). To scrutinize the data, and cluster the data independently on different sites a standard clustering algorithm would be executed. In other ways the centralized sites attempts to launch a global clustering based on local models, denoted as representatives. The local model creation might depend among position of object in various places which is not added for consideration. The gap to between the distributed and centralized cluster local model is much faster [1].

Load balancing is an essential problem in distribute clustering which is consider to be memory, CPU capability, network or interval load. To share work load between the different nodes of the distributed system is always required for better act of the system and to enhance the resource utilization. It helps to evade the condition of heavily or under loaded in network. The work load has been reduced using virtualization approach where server is created virtually and it does not seem as virtual to user and this diminishes the processing time, computation work and overloading in distributed clustering [2,3]. Limited issues of load balancing in distributed clustering are as follows, Throughput, Associated Overhead, Fault tolerant, and Migration time, Response time, Resource Utilization, Scalability and Performance. Therefore implementing a method should overcome above mentioned issues.

Need For Load Balancing

There are numerous nodes in a distributed structure. More number of tasks are arrived in distributed system are not arrived in a uniform distribution. Here some servers are heavily loaded where as some are lightly loaded [4]. If heavily loaded nodes are redistributed to lightly loaded nodes then it will increase the overall throughput of the system. This is known as load balancing. The objective of virtualization in distributed system is to enhance the performance of the system mainly it focuses on resource availability or response time based on distribution workload among the set of co-operative hosts. Instead of searching the content from main server the processing of query will takes place through virtualization approach [5].

Related Works

The web has becoming the primary reason for information distribution. Where its usage are increasing in various fields like commercial, academic, entertainment, marketing and this popularity results creates traffic flow in the retrieval of records from a huge database [6,7]. In current situation, web intelligence (WI) deals with the scientific exploration of new territories of the web. Therefore data mining has a lot of scopes in e-applications. The objective is retrieving the required content from hidden patterns for better performance. Here soft computing techniques were used to achieve web intelligence. For efficient resource utilization in distribution computing obtained by allowing load migration which improves the performance and efficiency of computing application through load balancing. Here mobile agent based approach can reduce the network traffic and enhance flexible load balancing mechanism [8]. If two or more computers, CPUs, hard drivers, networks or other resources workload is distributed eventually by load balancing manner.

Clustering is one of the approached in extracting meaning full or useful groups from a huge databases. Similar contents are assembled into single clusters therefore it avoids irrelevant content from a group and it increases exact required content according to user query [9]. Henceforth cluster study mainly used for various purpose like data compression or efficiency finding the nearest neighbors data. Clustering analysis used in various field such as psychology and other social sciences, biology[10,11], statistics, information retrieval, pattern recognition, machine learning [12], and data mining. Distributed clustering usage increases widely because it deals with huge and heterogeneous datasets, it cannot be grouped centrally. Moreover it aggregate local results that are obtained each site in such a way to generate a global models. The issue was the location is difficult, time absorbing and generate improper and indefinite global clustering therefore it produce incorrect knowledge. Here k-means algorithm based approach but it generates global clustering dynamically [13]. Therefore the aggregation is considered to produce final clusters in a compact and accurate hence it increases overall performance.

The suitable soft computing techniques, for issues such as fuzzy is apt for hold understands ability of incomplete data, varied media and patterns. Where neural networks suitable for non-parametric, efficient, robust and shows moral learning. Mixed media model were selected by genetic and uncertainty of data handled by rough sets [14, 15]. Dynamic feedback based techniques, a collaborative agent system [16] are castoff for processing time issues in load balancing. The feedback collected from the nodes reflects the heterogeneity of the ubiquitous distributed environment, and this forms the basis for forecasting the processing power of a particular node in the proceeding epoch. The survey describes the new arrivals, challenges and techniques to overwhelmed are discussed in [17, 18]. This section clearly describes the methods used in our proposed approach and issues related to it and methods that are proposed in existing are also discussed.

Implementation of Proposed System
This section describes implementation of VACC approach with an objective to overawe the issues. The application of distributed computing is increasing widely due to its wide range of features. But the issue in distributed clustering is load balancing and most common issue is retrieving required data from a huge database. Therefore these issues are taken into concentration and this work focuses on overcoming in it.

Server Virtualization
The masking of resources is called server virtualization. This includes total number of individual physical servers, processors and operating systems from server users. This works by dividing one physical layer into several isolated virtual environment which obtains by server side administrator software applications. This virtual environment is also identified as virtual private environments. Theses virtual private atmospheres otherwise called virtual private servers. The other names of representations like invitees, instances, ampules or emulations. Three vital approaches are follows in server virtualization: the virtual machine model and virtualization at the layer of operating system. Generally virtual machines are depends on the source and guest paradigm. The virtual imitation used to runs guest in hardware layer. Therefore this permits the invitee operating system to ride without any changes. If any administrator is using different operating system this approach allows creating guest. Therefore the guest has no knowledge of the host's working os because the running does not aware about the running on real hardware. Hence through virtual server environment the overloading of query handling would be handled easily and it would overcome the issues discussed previously. It diminishes the time consumption, query handling and overloading respectively.

Ant Colony Clustering
Once load balancing processing is done then extraction of data process should take into consideration. The efficient and effective data retrieval of information should be implemented for better processing. One of approaches in retrieving the data from a large database is ACC. Generally clustering will create groups based on a criteria however clustering results better performance. The working of ACC is described below. Swarm intelligence (SI) is a working of collective behavior of decentralized and self-organized system created either naturally or artificially. Ant colony clustering is one of methods in SI. This approach obtains better result in searching the content exactly from a huge database.

The ant colony clustering algorithm is described by the following pseudo code:

Step 1: Initialization, number of ants, entire number of iteration, and database size.
Step 2: Query is received from user and extracted from a huge database.
Step 3: Each ant is currently unloaded or consider being empty.
Step 4: Each ant is set a random speed S.
Step 5: Initially first iteration is inaugurated, based on criteria (Food source) cluster is formed. Similarly iteration is continued and optimal content are gathered into one cluster based on criteria.
Step 6: Graph structure will be twisted and it increases searching effectiveness and it reduces searching time.
End.

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Query is gathered from user and sends the query to database for efficient data retrieval. Iteration will be suggested established on the study of database size. Once size is analyzed initialization is done Ants A=0, Iteration I=0 and Speed of ant in searching source S=0. Number of iteration is identified n and suggested to I. Hence clustering is formed with deference to user query and result is obtained as output. A graph like structure is generated where it reduces searching time and produce better result in the constraints of efficiency and effectiveness. The cluster formation is shown in below figures.

Algorithm:
Data set components C={C_{ij}}
i=1,…..,n;
j=1,…...,|D_i|;
Parameters initially starts as S=0, i=o, A=0;
Iteration will be increased based on datasets;
N(i)⊆ C;
Graph construction is implemented in iteration process, G_c(V,E);
Graph construction is based on number of iteration G_c∈ N(i);
$p(c_{ij}|s)=\tau_{ij}^{\alpha} * \eta_{ij}^{\beta} / \sum C_{ij} \in N(S)\ \tau_{ij}^{\alpha} * \eta_{ij}^{\beta}, \forall c_{ij} \in N(S)$;
where
τij - pheromone value
ηij - heuristic value (desirability of edge) associated with the component *cij*.
Besides, α and β- positive real parameters whose values between pheromone versus heuristic information.

The complete working of planned system is shown above. In general, thousands of queries will be received per second. Where scheduling the mission is critical and difficult process. This causes the issues like traffic occurrence, reduction in performance, overloading and efficiency reduction. To overwhelmed these issues a load balancer process is implemented where this analyze server capacity and allocate the queries according to it. Once the queries are scheduled the grouping process will be done based on queries among the distributed computing. Ant colony clustering was implemented to retrieve results based on queries, and it forms a graph like structure where this reduces retrieving time absorption. The retrieved result will be displayed to users as result. Therefore it shows result obtained is in effective and efficient manner.

RESULT AND DISCUSSION

An educational dataset is taken for testing and working of our algorithm is implemented and results has been obtained are shown and discussed clearly below. Initially datasets are loaded and algorithm is assigned to it and processing is done. Hence the results demonstrate the variation among the algorithms and evaluate the accuracy among them respectively. The below result discusses the loading and implementation proposed approach and mode of testing's various approaches. Basic information and used scheme are listed below. Initially the time take to build model is 0 seconds. Here stratified cross validation is used and summary of it were listed below.

The above graph shows comparison of URL and accuracy values and various color in graph shows the number of instances takes place. X axis indicates URL and Y axis indicates Accuracy and the result obtained is plotted in the graph and shown above. The above graph shows comparison of title and accuracy value and various color in graph express the quantity of instances takes place. X axis indicates Title and Y axis indicates Accuracy and the result obtained is plotted in the graph and shown above. The X axis indicates the category and Y axis indicates the accuracy value. The plotted points in the graph indicate the number of instances occurred between these parameters. Various colors indicate total number of instance occurrence.

The relationship between accuracy and description is described above. Based on datasets the occurrence of instances is plotted in the graph has been shown clearly.

Performance Evaluation

In this section, the enactment of planned system is discussed and shown the way that projected method obtained better performance when associated to other. The parameters that are considered for evaluating performances are efficiency, effectiveness, load balancing and accuracy. Preexisting algorithms like Fuzzy, SVM, Evolutionary computing and proposed VAAC algorithms were compared with number of datasets in order to obtain load balancing. The load complementary is not the motive of soft computing algorithms is to retrieve efficient content from a large database. The proposed method shows better results in load balancing factor and retrieval of required content. The above graph describes the accuracy of required content in retrieval of data from a large datasets. Here proposed method retrieve content in iteration model where it increases accuracy in retrieved content. Here Y axis represents the percentage of accuracy and X axis represents the algorithms. The above graph shows that existing algorithm consumption to process a query and proposed method consumption to process a query. Here the results shows clearly the planned method obtain better results in efficiency, effectiveness and balancing the load.

CONCLUSION

In this work, VACC approach was proposed and implemented in order to overcome the issues faced in distributed clustering. However its advantage increases its application few issues affects its performance. One of the most vital problem is load balancing. This key factor alone leads this approach and it works by dint of virtualization, here server is created virtually and overloading processing where reduced and it attains better performance. This reduces overloading problem in processing queries and it is a crucial way to raise performance of the system. The retrieval of required content is implemented by Swarm intelligence in order to tackle difficulties in critical situation and it forms the clusters by iteration method which gather group's step by step process and this ensures accuracy in data retrieval from a vast database. Our approach obtains better result when associated to auxiliary existing tactics are shown clearly and it obtains well performance.

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Table 1: Scheme and Test Mode Execution

SCHEME	RELATION	INSTANCES	ATTRIBUTES	TEST MODE
Naive Bayes	Query Result	30	5	10 fold cross validation
Simple K means	Query Result	30	5	evaluate the training data
Filtered Associator	Query Result	30	5	Associator model (full training set)
Cfs Sub set Eval	Query Result	30	5	evaluate on all training data

Table 2: Accuracy Measures

Kappa statistic	0.0943
Mean absolute	0.1021
Root mean	0.2301
Relative absolute	92.5432 %
Root relative	97.464 %
Total Number of	30

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Table 3: Evaluation of accuracy and attribute selection

Data set	Algorithm	Accuracy	Attribute selection
50	Fuzzy	42.8571 %	4
50	SVM	52.4231 %	3
50	VACC	74.2487 %	2
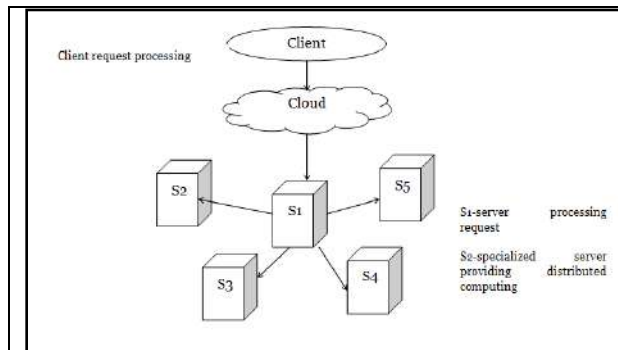
Figure 1: Distributed Clustering Architecture
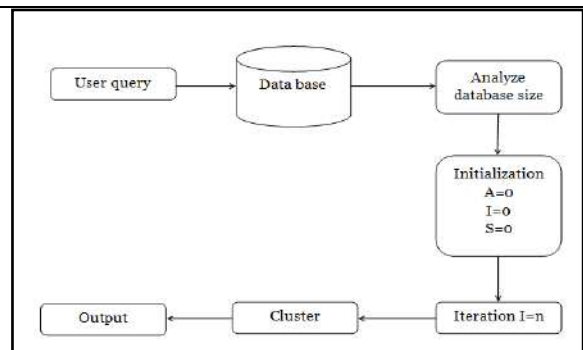
Figure 2: working of ACC
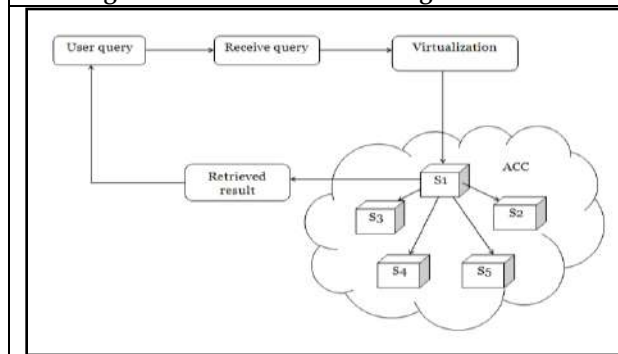
Figure 3: Overall working of projected approach
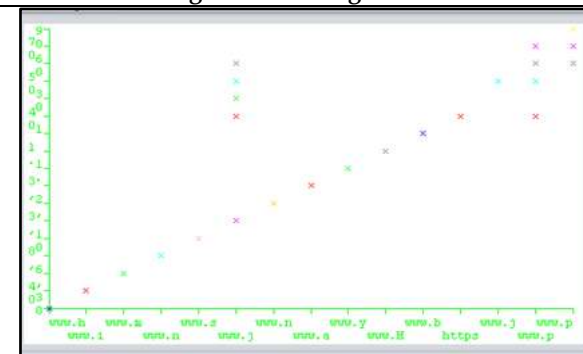
Figure 4: comparison graph between URL and accuracy value
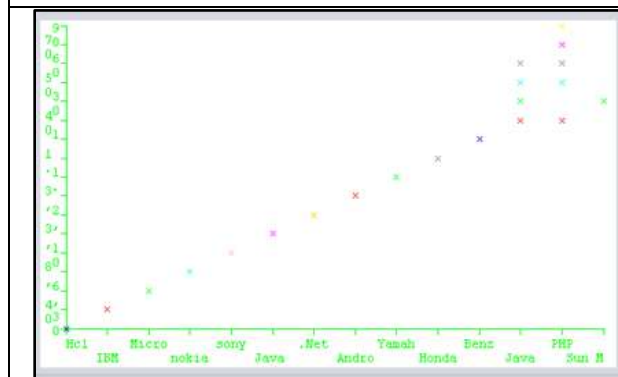
Figure 5: comparison graph between title and accuracy value
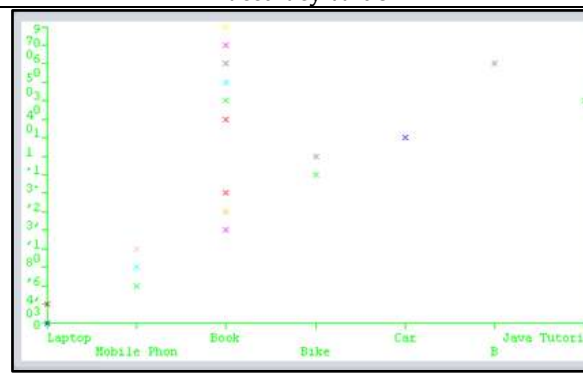
Figure 6: comparison graph between category and accuracy value
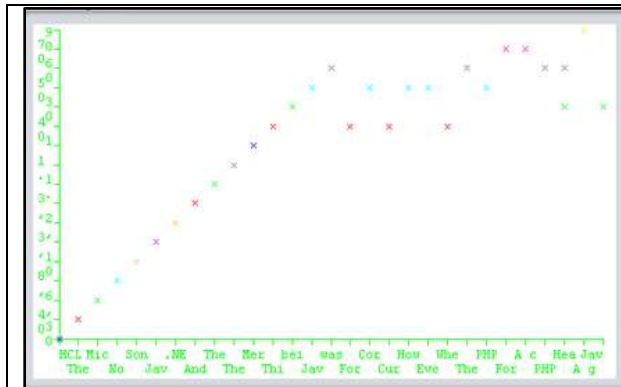
Figure 7: comparison graph between description and accuracy
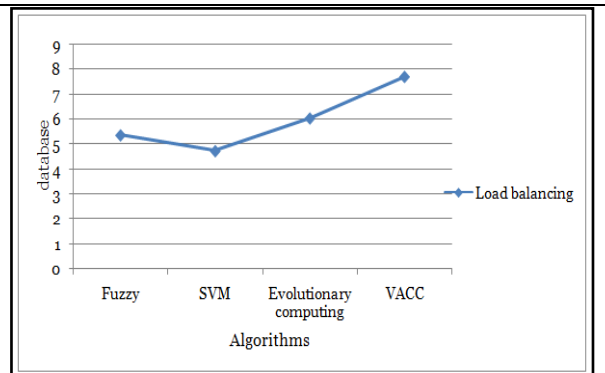
Figure 8: Comparison of algorithms vs load balancing
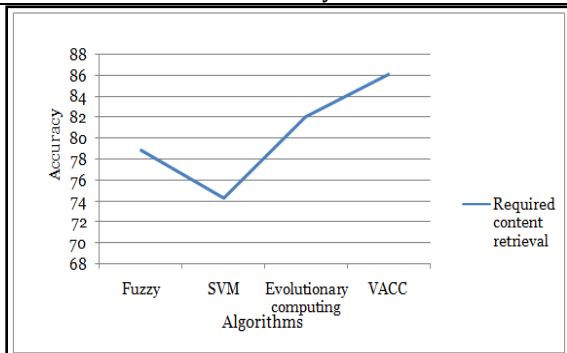
Figure 9: Comparison of algorithms vs effectiveness
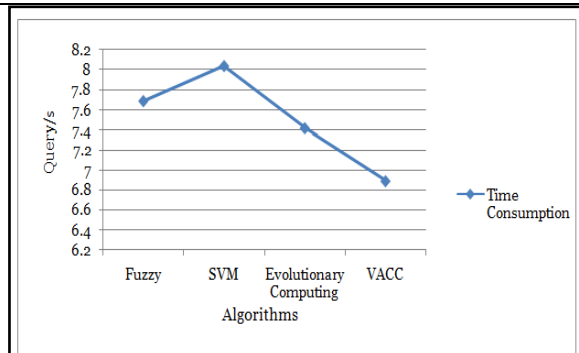
Figure 10: Time consumption of proposed method

RESEARCH ARTICLE

Ensemble Learning Regression Models for Fatigue Life Estimation

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ABSTRACT

The extensive use of sandwich composites is seen in automotive, marine, and civil constructions. In most applications, the sandwich composites encounter cyclical loading, which leads to failure. As the composites are made by stacking- up the fiber layers, it becomes obvious to study the fatigue behavior of these layers during use. Therefore, a thorough knowledge of laminate behaviour under cyclical loading can result in better material design for fatigue applications. Fatigue failure is a incredibly common failure among most of the composites and is not well- understood by materials engineers. Even now, material designers are depending on the physical testing of these materials before putting it to use. These physical testing's cost time and energy during the development stage. It is thus the ideal platform to apply machine learning (ML) on the key estimates of the fatigue behavior of the sandwich composites. Ensemble learners are employed to estimate the product's life cycle rather than single ML models.

Keywords: Composites, Fatigue failure, Machine learning, Ensemble learners.

INTRODUCTION

Automotive, military, aerospace, and other domestic product producers are looking for newer and better materials to increase efficiency and performance throughout the product lifetime. Every alteration made during the product design process is aimed at improving performance. The use of composites results in properties such as reduced weight and increased bending strength. In terms of the sandwich class, composite materials have expanded the scope of structural applications. Some specific applications of engineering demands tailor-made properties in completing the material design. The optimal design process must be envisioned through careful composition selection and appropriate production procedures. In composite material science, better materials are obtained through a better understanding of the constituent materials, i.e., better correlation in terms of qualities like modulus, stress, and load bearing capabilities. Every understanding of inter-constituent relationship is very critical during the composite material design. The above task can generally be completed by developing a mathematical modeling derived from experiments [1].   It's past time to invest in materials that will allow you to change your design and develop. As a result, a novel approach to management is necessary in order to obtain an optimum sandwich composite. Sandwich

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composites have a higher strength-to-weight ratio, bending strength, and stiffness than other types of composites. To generate anisotropic outer surface frameworks, several new sandwich systems use carbon and Kevlar fibre reinforced polymers (composite materials). [2]. Such materials allow greater control of the horizontal resistance characteristics and the design stability of the final products compared with conventional metals and alloys with isotropic characteristics. Polymer matrix composites are critical to improving product reliability and economy in industries such as automotive, military, and aerospace, where manufacturers are looking for newer and better materials to extend the lifecycle of their products. The majority of businesses are investing extensively in the development of new materials that can improve performance [3].

The literature supports the dynamic problems caused in sandwich composites working with low cyclic or high cyclic stress amplitudes. Fatigue failures are responsible for 80% of accidents, according to numerous authors [4]. Therefore, it is obvious that it is essential to study fatigue response and failure mechanism of these materials. Many authors have worked on the characterization of honeycomb structures. Researchers investigating the influence of fatigue life of sandwich composites using finite element techniques considered a few defects connected with the fatigue behaviour. The analysis was used to find out the mechanical parameters and the fatigue life of honeycomb sandwich structures with flaws [5]. The material resistance during fatigue behaviour determines the structural sandwich composite's reliability. Sandwich composites with colourful cores and fibres were chosen for this study, and the reason for fatigue failure in terms of fatigue cycles was investigated. Figure 1 demonstrates a wide range of sandwich composite failures [6]. In order to make superior sandwich composites, the correlation between the internal and hyper properties is critical. Before fabricating sandwich composites for appropriate applications, it's critical to understand all of the properties [2]. For fatigue applications, sandwich constructions are not as well understood as metals. During fatigue loading, sandwich composites displayed observable mechanical deterioration characteristics. Delamination, fibre breakage and debonding, substrate splitting, and interfacial fibre/matrix debonding are the most common failure features [7].

Predicting fatigue behaviour is always hindered by the loading complexities in many composites. Fibres in composites always pose problems for better fatigue life. To understand the behaviour of any composite during application, a real-time understanding of fibre behaviour is required. As a result, extensive testing of composite fatigue life is advised, which adds expense and time [8]. These intensive testing and examinations lead to a quantity of different newer experiments having too many varying factors. The problem is also compounded by the emergence of new materials needing assessment and rising complex attributes for these products. Moreover, a designer must be able to resolve the fatigue response of either a newly developed or in-service structure in active condition to find the best solution. As scarce fatigue data is available in the materials repository, it is obvious that the test data and trained data must be compared over a small repository of fatigue test data [9]. Therefore, finding fatigue lives for a sandwich composite tends to a system, which promotes the designer to imply initial predictions on fatigue life using the data repository. In recent times, most material engineers affirm the materials for reliability using Artificial Intelligence and machine learning techniques.

Data mining or machine learning acts as a key role in the prediction of materials properties. Using the sample dataset, one can automatically learn the features and work on large linear and nonlinear datasets. As Several classifiers or regression models combined together is called ensemble learning. It gives a model, which is more perfect and reliable. In materials research, it is necessary that both the methods should be capable of merging machine learning and statistical methods [10]. Tree-based approaches not only produce high precision rates, but also enable analysis of the significance of the attributes used in the predictions [10]. In fatigue lifecycle predictions, various hierarchical factors, which effect are core shear strength, face bending strength, fatigue stiffness, and fatigue cycles to failure. Tree based learning requires minimum preprocessing data and skill to fit the nonlinear data in useable format [11]. As a consequence, there is an increasing need to test tree- prediction approaches and equate them to alternative approaches. To this end, a rigorous assessment is desired for a clear understanding of the potential of tree-based ensemble learning to predict fatigue lifecycle. The intention of the present work is to evaluation the fatigue lifecycle of SWCs using ensemble learning methods.

DATA AND METHODS

Data Description

The sandwich composites dataset used in the present work was based on the fatigue experiments conducted by Manujesh et al. [12] with 3-point contact fatigue as shown in Table 1. Four different varieties of sandwich composites were fabricated comprising of E-Glass fibres with change in PU foam densities. The material variety used in the present study had varied stacking sequence and differed with the matrix and fibre volume fractions. The data of 1080 samples was used to predict the fatigue life of the sandwich composites. Test parameters like face bending strength and core shear strength along with flexural and shear modulus against the fatigue cycle was predicted. The fibre volume fraction ratio considered was 60 %, 65%, and 70% along with three different stack-up sequences such as ±0 [±30,±45]₃s, ±0 [±60,±95]₃s and ±0 [±0, ±30, ±45, ±60, ±90]₂s. For all the combinations of the aforementioned sandwich composites, the fatigue test data collected was checked analytically. Later, these data sets were used to predict statistically using machine learning methods. Though the experimental data used was accurate, yet the analytical data too can reach the same approximation with more costly approaches. The machine learning method was used for decision-making in building different types of sandwich composites [13]. The present work is carried out based on the data set obtained from the experimental study of Rao, R. V. and Manujesh B. J[14] .

Fatigue Data Pre-Processing

The data was collected from experimental and simulation studies. Around 1080 sample data was used in the present study. The following pre-processing methods were performed over the experimental data using appropriate regression analysis models:

- The values were normalized in [0, 1] for layup sequence 'θ',
- Maximum bending strength 'σ_{max}' and flexural modulus 'σ_a', and
- Calculated and used logarithmic values and cycles to failure (N).

This was achieved because 'N' requires loading from 500 to 5×10^5 cycles. Training a neural network with such a large variety of values would result in incredibly low results in the simulations. The experimental fatigue data is tabulated in the Table 1.

Methods for Regression

The sections below give a concise prologue to different tree-based learning algorithms, specifically individual and ensemble algorithms is given.

Regression Tree

It is like a decision tree. The Regression Trees main objective is to locate homogeneous classes through space division using minimization algorithms. Here, model building involves two steps, that is, tree construction and pruning [15] . Without considering outlier and generalization, the tree has to be constructed. In the trimming phase, the complete-grown tree is pruned to eliminate branches that reduce the generalization potential. Only after training of regression can the structure of the tree be determined. For predicting a new instance, it starts for the root, and possibly all the branches for which it satisfies the condition, until the leaf node. Then prediction is made based on the mean value of all the data records in that specific leaf node [16].Figure 2 shows the regression tree representation.

Bagging

Bagging is also called bootstrap aggregator, and building a model here is by using the same learning algorithm, but each learner is trained on different sets of data. In statistical classification and regression, these are used to develop the consistency and accuracy of machine learning algorithm. It also eliminates uncertainty and tends to prevent over-fitting [17]. For supervised learning, bagging and boosting are designed. The goal is to build many sub-sets of data from a randomly selected training sample with a substitution. Each array of subset data is used to train the decision trees. Therefore, different model ensembles are required. To give a robust and better model, the average of all the

predictions from various trees are calculated.   From the original dataset, the data has to be collected randomly by replacement. Some of the 'D's in a sample bootstrap 'D1' may be held numerous times, and the rest is excluded. The bootstrap principle states that sampling from a raw dataset ('D') is the best approximation for sampling from an unknown distribution ('P'). This so-called bootstrap sample is then used to build the model. The process must be repeated a total of 'N' times, resulting in N models. Figure 3 depicts the bagging structure employed in the current study.

The final combination of bagging and boosting are linear combinations of the regression models. When several regression models have been built, the average assessment of the consequences of each regression model shows the final prediction [18].

Gradient Boosting Regression Tree

Boosting is a significant collection of machine learning approaches that differ slightly from bagging. The common technique of boosting is to create a sequence of models where each model is skilled on a re- weighted sample of the original data collection. Sequentially the models are trained and then the errors are estimated [19]. The aim is to improve the precision of the previous model. The model is retrained each time by adjusting its weights [20]. This process converts weaker models to stronger ones. Gradient boosting collects the subsamples by without replacing the data in every iteration. To evaluate the model update for new iteration, the sub-sample is employed instead of the original data.

Random Forest

It is an ensemble method of regression. It is a bagging approach, and not a boosting. Since it is used for regression analysis, it is also called regression forest. It is destined for nonlinear multiple regressions. Every path from the leaf leads to the allotment of a continuous variable [21]. Here, variance of the ensemble is designed using Equation 1.

$$\text{var} = \rho\sigma^2 + \frac{1-\rho}{B}\sigma^2, \tag{1}$$

Where,
B= No of trees,
P= Pair -wise correlation between the trees, and
σ2= Variance of each tree.
Random forest adjusts the code to the way sub-trees are trained so that subsequent projections from all the sub-trees have less association. It is an easy adjustment. Within CART(Classification and Regression Tree), when choosing a split point, the learning algorithm examines all the attributes and all equation values within order to choose the most suitable split point. A random forest algorithm adjusts this process, so that the learning algorithm is restricted to a random sample of features to be scanned [22]. For balancing the opposing factors, the variance has to be reduced. Rather than the sample data, the use of the full training data is invoked to decrease model bias.

Model Evaluations

Generally, for predictive models, the following performance measures are used. The correlation coefficient (R) is a common indicator of how the curve corresponds to the raw data. If there is an exact fit between the desired and actual values, then the value will be one, which means the same tendency value [1]. The numerical equation is:

$$R^2 = \left(\frac{n\sum y.y' - (\sum y)(\sum y')}{\sqrt{(\sum y^2) - (\sum y)^2}\sqrt{(\sum y'^2) - (\sum y')^2}}\right)^2 \tag{2}$$

Here, y=real value, y'=predicted value, and the number of data samples is n.

The square root of mean square error is measured by root mean squared error (RMSE). The RMSE is therefore, the average distance between the data point and the installed line determined along the vertical line [14]. To obtain RMSE, the equation is:

$$RMSE = \sqrt{\frac{\sum (y' - y)^2}{n}} \qquad (3)$$

Mean Absolute Error (MAE) is the sum used to calculate, how close the predictions or assumptions are to the final consequences [11]. And it is given by,

$$MAE = \frac{1}{n} \sum_{i=1,n} |y - y'| \qquad (4)$$

In case the amount of data is small, then cross-validation is required to prevent from over-fitting. Here,the data is divided into fixed number of partitions and analysis is executed on each partition, and then finally,the average of the overall error estimation is calculated . In this process, the data is divided in to two parts, one for learning the model and the other to validate the model. The learning and validation sets will run-over in consecutive iterations in such a manner that each data point has the probability of being tested against each other for standard cross- validation. The K-fold cross- validation is one of the best choices when handling small datasets [23]. Here, the datasets are spilt into K separate sets t_h(h=1,2,…..K) of around the same size. K times the model is trained, and each time when it is being trained, it will leave one of its samples. It uses only missing sub-sets to measure the error of the forecast. The average value of sub-set of K values is the cross- validation approximation of the added sample error. Various folds of cross-validation are used to assess the performance of the predictive model. The same fixed datasets are randomly divided in to 'k' partitions. Every time, out of all partitions, one portion is held out, which learn the model for k-1 portions. By using the hold out, the set error is calculated.

RESULTS AND DISCUSSION

The goal of this research is to assess the performance of various tree-based learning methods, including regression tree, random forest, and gradient boosting regression tree. The model is built with the Python sklearn library package. The independent variables utilised to learn the model are volume fraction, core density, layup sequence, face banding strength, flexural modulus, shear strength, and shear modulus in this study. The correlation coefficient 'R' is used to calculate model fit and predictive performance following stratified five-fold cross-validation. The outcomes of several tree-based regression algorithms are examined in this paper. The statistical findings of the various algorithms utilised in the study are shown in Table 2. It describes the results obtained utilizing ensemble methods and individual regression trees, random forests, and gradient boosting algorithms (i.e., bagged regression tree, bagged random forest, and bagged gradient boosting regression tree). The suggested predictive models' efficiency is determined using mean absolute error (MAE), root mean squared error (RMSE), and correlation coefficient R output statistics [24].

Only the data from the test sample is used to determine the prediction's accuracy. The accuracy of the prediction is measured by a good learning (training) sample, but it does not guarantee the accuracy of the prediction. In recent years, ensemble machine learning approaches have gotten a lot of attention, and they've led to significant improvements in regression problem results. The first empirical study results ('K' value for cross-validation is 5) are given in Table 1. It has noticed that, when considering a Bag, the GBRT model has the best performance in case of 'R' (0.9676), and the next best model is the single GBTR with 0.9345 as the 'R' value. From the table, it is observed that single regression tree yields worst performance compared with all the models. In addition, MAE and RMSE statistics are in-consistent with the correlation coefficients given in figure 4.

Sanjay Mathur *et al.* [25] used ANN for predicting the fatigue life of carbon fibre reinforced plastic and found correlation coefficient of 0.865. Jui-Sheng et al. designed two ensembles of decision trees and found that the bagged

regression methods yield0.8904, 'R' value, which performs better compared with single learning models [26]. In the present work, individual learning and testing, the best performance with the highest 'R' value for GBRT was found to be 0.9345. While the mean square error and root mean square error was inconsistent with the purpose of correlation statistics. Other parts of the table affirm that the ensemble method always yields better performance with the single model method. For the present study, two empiric studies were initiated using a training-testing method. The 5-fold cross-validation was applied to find the performance of the regression model. The split sample method is a useful technique for determining the quantitative quality of a test set. In the split sample process, the dataset subsample is held back from training and used to test the precision of the forecasting [27].

By using the test dataset, optimum individual regressor and ensemble regressor was performed. Notably, the comparison was made with one randomized training-test segment, which had comparable predictive success on the training set with hyper parameter tuning tests. Figure 5 illustrates the comparison of the correlation coefficient between the actual and predicted value of fatigue life.

CONCLUSIONS

This research provides the first systematic assessment of the efficiency of tree-based ensemble learning algorithms for fatigue life prediction in sandwich composites. Prediction of fatigue life of composite material is affected by various factors such as stress amplitude, stress ratio, monotonic properties, fraction of fiber volume, etc. The statistical distribution of fatigue life data also affects the prediction of fatigue life. Here, ensemble learning algorithms such as random forest, regression tree, and GBRT were studied. To optimize predictive accuracy and reduce the variance, ensemble methods are preferred. Fatigue lifecycles with different models were estimated and compared. The performance measures revealed that for prediction of fatigue lifecycle, the ensemble methods work really well. Highest accuracy can be obtained by the ensemble GBRT method. As a result, it can be concluded that ensemble methods are prominent technique for fatigue life prediction.

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Table 1: Experimental Fatigue Data

Sl. No.	Material Type	Volume Fraction (%)	Core Density (kg/m³)	Layup Sequence (θ)	Face Bending Strength (σ_{max}, MPa)	Shear Strength (MPa)	Shear Modulus (MPa)	Fatigue Lifecycle (N)
1.	CSM-PUF-CSM	60	100	0,90; ±30;±45; ±60; ±90	1.46	0.092	0.810	276243
2.			200	0,90;±30;±45; ±60; ±90	1.59	0.095	0.971	293271
3.	CSM-S-PUF-CSM-S	60	100	0,90;±30;±45; ±60; ±90	1.43	0.09	0.81	266704
4.			200	0,90;±30;±45; ±60; ±90	1.64	1.03	0.976	284327
5.	WR-PUF-WR	60	100	0,90;±30;±45; ±60; ±90	1.54	0.113	0.814	263593
6.			200	0,90;±30;±45; ±60; ±90	1.69	0.118	0.976	291794
7.	SBM-PUF-SBM	60	100	0,90;±30;±45; ±60; ±90	1.44	0.094	0.81	298711
8.			200	0,90;±30;±45; ±60; ±90	1.47	0.099	0.99	326327
9.	CSM-PUF-CM-S	60	100	0,90;±30;±45; ±60; ±90	1.49	0.09	0.81	293516
10.			200	0,90;±30;±45; ±60; ±90	1.58	0.094	0.867	295732
11.	CSM-PUF-WR	60	100	0,90;±30;±45; ±60; ±90	1.48	0.091	0.81	309111
12.			200	0,90;±30;±45; ±60; ±90	1.59	0.114	0.974	327319
13.	CSM-PUF-SBM	60	100	0,90;±30;±45; ±60; ±90	1.45	0.09	0.81	316739

Table 2: Performance statistics of different regression methods

	Single models			Ensemble models		
	RT	**RF**	**GBRT**	**Bag-RT**	**Bag-RF**	**Bag-GBRT**
R	0.708	0.823	0.934	0.725	0.864	0.967
MAE	4.06	4.56	4.43	4.26	4.34	3.30
RMSE	5.57	5.120	5.07	5.43	4.98	4.08
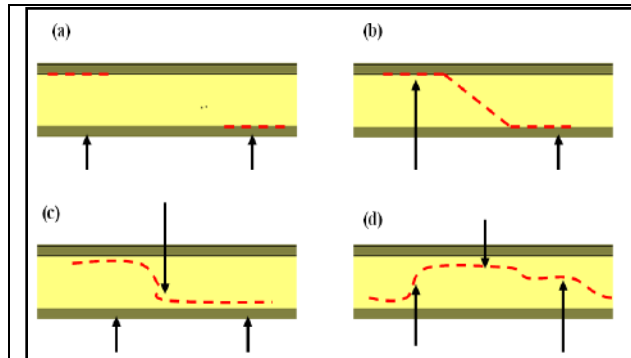
Figure 1: Failure modes of low density sandwich composites under fatigue loading - a) Low density/low frequency at lower cycles, b) Low density /low frequency at higher cycles, c) Low density/higher frequency at lower cycles, and d) Low density/higher frequency at higher cycles
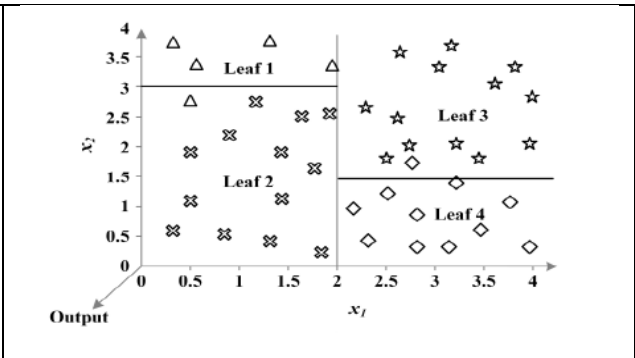
Figure 2: Regression tree sample
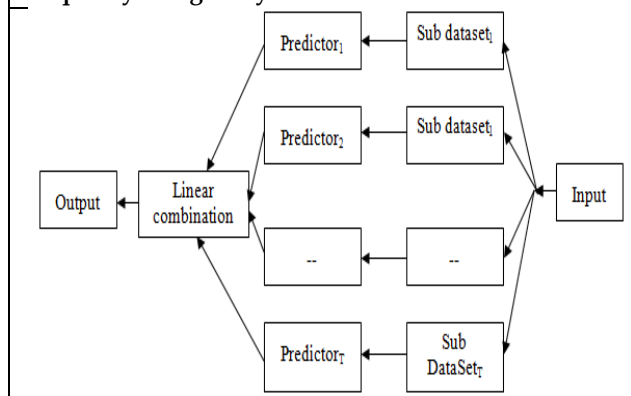
Figure 3:   Model Structure of Ensemble Method (Bagging)
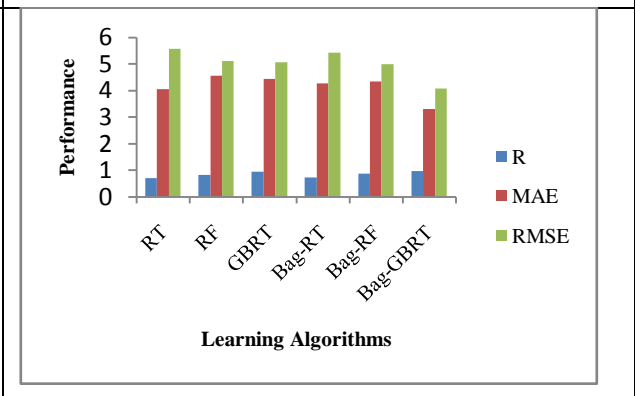
Figure 4:   Comparison of Learning Algorithms in Terms of Various Performance Measures
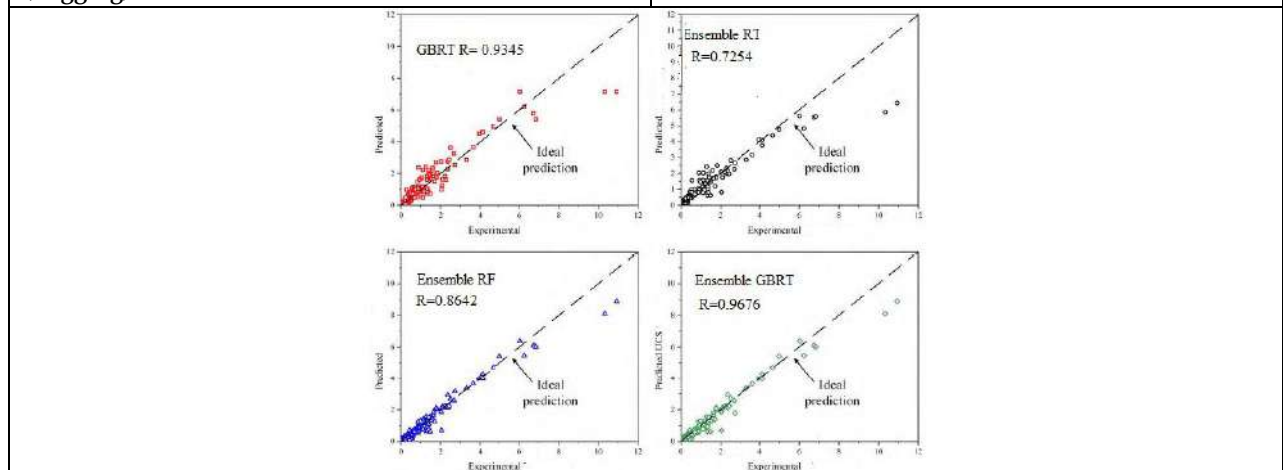
Figure 5: The correlation coefficient value R for test data (i.e., Individual Regressor and ensemble Regressor)

RESEARCH ARTICLE

Crop Yield Prediction using Machine Learning Models and Neural Networks

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ABSTRACT

Climate change, combined with population and wealth increase, poses a challenge to the agricultural industry and global food security. This makes it difficult for the famers to estimate the yield. Farmers would benefit from being able to predict crop yields ahead of time so that they make suitable crop production measures. To predict the crop yield there are many different models available. In this paper we used Machine learning and neural network models to examine which algorithm performs better. Models used are - Random Forest, XGBOOST, Back Propagation Neural Network and ANN for prediction of crop yield. The paper focuses on few of the seasonal, annual and perennial crops grown in all 14 districts of Kerala. The data contains crop data along with it's weather parameters. Four different metrics were used to evaluate performance of the models. XGBOOST followed by Random Forest performed better. In conclusion, Machine learning models outrun Deep learning models.

Keywords: Random Forest, XGBOOST, ANN, BPNN, RMSE, MAE, MAPE.

INTRODUCTION

Agriculture is crucial to India's economy. India is the second-largest agricultural product producer in the world. For around 58 percent of India's population, it is their principal source of income. Weather conditions, soil characteristics, and crop area allocation are all important factors in agricultural productivity. Yield prediction aids farmers in making the best use of their increasing crops. Yield forecasting provides a clear picture of what to expect in the future. Kerala's agriculture industry has been having difficulty expanding. Agriculture and related industries (including crop, livestock, forestry and logging, fishing and aquaculture) experienced an annual growth rate (GSVA at constant 2011-12 prices) of (-)6.31 percent in 2013-14, 0.02 percent in 2014-15, (-)5.10 percent in 2015-16, and (-)0.65 percent in 2016-17, according to data from the Directorate of Economics and Statistics (DES). In the 2017-18 fiscal

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year, the industry grew 1.72 percent. However, in 2018-19, growth fell to (-)0.52%. Hence, predicting yield not only helps the farmers for management of crops but also helps in preventing famine, particularly in an era of global climate change [1]. It is also being able to estimate the ability to produce enough food to meet human needs. There are enormous factors that needs to be taken into consideration for yield prediction. Climatic condition and weather play a huge role in crop production and these factors may vary in different regions, by analysing these factors there isn't a proper model introduced as all these factors are extremely different in different regions of India. Kerala has an agrarian economy, with a network of blue backwaters, rivers, and streams. Agriculture is facilitated to a large extent by the amount of water provided by 34 lakes and various tiny stream-lets and waterbodies, as well as the state's ample annual rainfall of 3000mm. As a result, agriculture dominates the state's economy. Paddy, coconut, and plantation crops such as rubber and banana are the main crops farmed in Kerala.

In research focused on paddy yield prediction using MLR-ANN, a hybrid algorithm. Thirty years of paddy data were provided by the state of Tamil Nadu's department of economics and statistics. A total of 745 cases with 16 attributes were modelled. Pre-processing was done before feature extraction, and the retrieved features from the feature extraction model were input into the model. MLR intercept and coefficients were used to initialise the ANN's input layer weights and bias, the proposed hybrid model was used to analyse prediction accuracy. A comparison models is conducted using performance metrics [2]. Researchers applied random forest approach to predict yield. Climate and soil factors such as rainfall, temperature, humidity, soil quality, and so on are taken into account in the data set collected from the respective agricultural department. After pre-processing, they used the RF (Random Forest) and then provided input parameters such as District Name, Crop Name, Area, and Soil Type to forecast the yield[3]. In a paper which tested the prediction accuracy of machine learning algorithms for crop production prediction in ten crop datasets using data from an irrigation zone in Mexico. Various machine learning approaches were examined. The RMSE, root relative square error (RRSE), mean absolute error (MAE), and correlation factor were used to verify the models. KNN has the lowest average RMSE, RRSE, MAE errors and greatest correlation factors in M5-Prime, making it an ideal tool for agricultural planning [4].

Various machine learningapproaches were used to forecast crop productivity (RNN, LSTM, RF, XGBOOST, KNN, ANN, logistic regression, linear regression). Temperature, location, rainfall, and season are all taken into account. The names of fifteen commonly produced crops in India were predicted based on the characteristics. On the dataset, various classifiers were trained, and a confusion matrix was plotted, as well as comparisons of various models [5]. In research focusing on forecasting rice yield using a Support Vector Regression model with multiple kernels for the entire country and the top five paddy-producing states. Independent variables such as area and production were used. Cross-validation and hyper parameter optimization of numerous kernel parameters were used to choose the best fit models. The training and testing dataset's RMSE and MAE were determined [6]. An approach by splitting into two phases, training and testing was implemented. The pre-processed data is clustered using the K-means algorithm, and then association rule mining is performed on the data clustered, with the rules derived using frequent pattern mining. The yield is anticipated based on the rules once the district and crop are entered into the prediction model [7]. Prediction of rice yield using a weather-based system was done. Agricultural data, rainfall, humidity, and temperature are all taken into account. First, using neural networks, a model was created to predict meteorological parameters, and then rice yield was evaluated using SVR, which takes into account both projected weather and existing agricultural data [8]. Artificial neural network with Feed Forward Back Propagation was employed for prediction of yield. Neurons are only connected forward in this network; there are no connections to the back, in a back propagation neural network, however, neurons are linked backward. For a given input, the predicted and actual outputs are compared. Predicted outcomes are used to adjust the weights of the individual layers backward from the output layer to input layer in back propagation training. Each neural network layer is linked to the layer above it. Based on a range of predictor criteria, the methodology was utilised to estimate and forecast agricultural yields. We looked at ANNs with zero, one, and two hidden layers. MSEs have been used to determine the optimal number of hidden layers as well as the ideal number of units in each hidden layer. It was then analysed using a MATLAB to improve its efficiency[9].

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The crop yield is predicted using a Random Forest classifier. During the training stage, a large number of trees are created, and the output is split into categories depending on the number of classes (classification), and class prediction (regression). Only two-thirds of the dataset is considered, with the rest being utilised as a test bed [10]. Wheat, maize, and potato data was used for predicting respective yields. Data was collected from three different sources and employed MLR and random forest models on the data. Random forest outperformed MLR benchmarks and was determined to be extremely competent of predicting crop yields. In all test instances, the root mean square errors (RMSE) for RF models varied from 6 to 14 percent of the average observed yield, whereas these values for MLR models ranged from 14 percent to 49 percent [11]. Yield prediction can be done using different techniques and methods. In this paper we used different machine learning models and deep learning models to predict the crop yield and compare in terms of its RMSE, MSE, MAPE values. In this paper we are going to predict the yield of Areca nut, Tapioca, Coconut and Banana.

PRELIMINARY

Random Forest

Random Forest is a regression supervised learning ensemble learning approach. Ensemble learning brings together predictions from multiple machine learning algorithms to get a more accurate result than a single model. Random forest builds decision trees on different samples and also splits the features at every step randomly and takes their majority vote into consideration. Simply Random Forest can be expressed as Random forest= DT(base learner)+ bagging(Row sampling with replacement)+ feature bagging(column sampling) + aggregation(mean/median, majority vote) [12]

Mathematically it can be expressed as

$$\hat{f}_{rf}^B(x) = \frac{1}{B}\sum_{b=1}^{B} T_b(x) \qquad (1)$$

Here Tb is the random-forest tree for the bootstrapped data b; for b = 1 to B [19].

XGBOOST

XGBOOST stands for extreme Gradient Boosting. The gradient boosting decision tree approach is implemented by XGBOOST; it is an ensemble technique in which new models are added to rectify errors committed by previous models. Models are added in a logical order until no further enhancements are needed. XGBOOST avoids over fitting by taking care of missing values, tree pruning and parallel processing. XGBOOST uses gradient descent algorithm to boost weak learners.

$$F(X) = H0(X) + eta\big(H1(X)\big) + eta\big(H2(X)\big) \quad (2)$$

Where F(X) is the prediction from XGBOOST model, eta is the learning rate and H(X) is the second order derivative of the loss at current estimate [16].

ANN

The artificial neural network (ANN) is a computational model that simulates the function of nerve cells in the brain. An artificial neural network (ANN) is a type of learning algorithm that may adapt or train on its own in response to fresh input. As a result, they're a great way to model nonlinear statistical data. It consists of three components: input layer, hidden layer and output layer. Input layers takes inputs in different format, hidden layer performs the calculations to find the hidden features and patterns and output layer finally gives the result. The weighted total of inputs is computed using ANN. A transfer function is used to express this calculation

$$\sum_{i=1}^{n} g_i(w_i x_i + b) \qquad (3)$$

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Here I is the number of hidden layer, g_i is the activation function, W_i are weights, X_i are the inputs we pass into the model and b is the bias [17]. Weighted total is passed as an input to the activation function. The activation function used for predicting crop yield in this paper is *RELU* activation function.

BPNN

Back propagation is the foundation of neural network training. It's a method for fine-tuning neural network weights based on the error rate of the previous epoch. You can lower error rates and increase the model's generalization by fine-tuning the weights.

For each l = 2,3,…,L, we compute

$$z^l = W^l a^{1-l} + b^l, a^l = \sigma(z^l) \qquad (4)$$

Computing the error
$$\delta^L = \nabla ac\theta\sigma'(z^l) \qquad (5)$$

Backpropagating the error: For each l = L-1, L-2, …...
Compute
$$\delta^l = ((\omega^{l+1})^T \delta^{l+1})\theta\sigma'(z^l) \qquad (6)$$
Here W^L, b^L is weight and bias of the Lth layer [17].

METHODOLOGY

Study Area
All 14 districts of Kerala i.e., Alappuzha, Ernakulam, Idukki, Kannur, Kasaragod, Kollam, Kottayam, Kozhikode, Malappuram, Palakkad, Wayanad, Thrissur, Pathanamthitta, and Thiruvananthapuram were considered in this paper. The amount of land used for agriculture and production varies by district.

Data Collection
Since we are predicting the yield for district level in this paper, we acquired climate parameters for respective districts along with crop data. The Kerala Department of Economics and Statistics provided crop data such as production, acreage, and yield for all districts. The attributes given as input parameters are District, Crop, Season, Area, Production, Rainfall, Minimum temperature, Maximum temperature Humidity, Minimum wind speed, Maximum wind speed, precipitation and Yield. We acquired data of four different crops namely Tapioca, Coconut, Banana, and Arecanut. Past twenty years of data (2000 - 2019) of four different crops were collected. The dataset consists of 1120 rows with 13 variables. Basic pre-processing was done in the initial stage of data collection as the data were manually collected from different sectors of agricultural department.

Implementation
Firstly, the dataset was split into different crops for modelling. Data cleaning like checking for missing values and changing the data types is done. Through VIF (variance inflation factor) we observe that there is multicollinearity in the data. As we know multicollinearity has effect on model performance, the solution for this is to either remove variables which have multicollinearity or chose models that are immune to multicollinearity. Since, the percentage of variables with multicollinearity is high we chose models which are immune to multicollinearity. As shown in Fig 1 crop and climate parameters were collected and were combined together then the data was normalized to improve the model performance and the data was split into train and test. In the next step models- Random Forest, Artificial Neural Networks, Back Propagation Neural Network and XGBOOST were implemented and the performance of the models were evaluated using three different metrics *RMSE, MAPE,* and *MAE.* Later the results of these metrics were compared to evaluate the performance of the model. In order to interpret which model performed better.

RESULTS

Evaluating Metrics

To evaluate the models, the *RMSE, MAPE, MAE* values are used. The RMSE is the square root of the residuals' variance. It shows how well the model fits the data in terms of absolute fit–how near the observed data points are to the model's predicted values. RMSE value is calculated as

$$RMSE = \sqrt{\frac{1}{n}\sum_{i=1}^{n}(s_i - o_i)^2} \qquad (6)$$

Where, O_i are the observations, S_i predicted values of a variable, and n is the number of observations[13]. MAPE also known as mean absolute percentage error is used to measure forecast accuracy. It is basically the average of absolute percentage errors. MAPE is calculated as

$$MAPE = \frac{1}{n}\sum_{t=1}^{n}\left|\frac{A_t - F_t}{A_t}\right| \qquad (7)$$

Where, n is the number of times the summation iteration happens, A_t is the actual value, F_t is the forecast value[14]. Mean absolute error (MAE) calculates the average difference between the calculated values and the actual values. MAE is calculated as follows

$$MAE = \left(\frac{1}{n}\right) * \sum |y_i - x_i| \qquad (8)$$

Where, y_i actual value for the ith observation, x_i is the calculated value for the ith observation and n is the total number of observations [15].

RESULTS

The RMSE, MAPE, MAE values for different models were shown in the table 1, table 2 and table 3 respectively. By table 1, we observe that RMSE value was least for random forest model ranging from 0.015 to 0.037. The less the RMSE value the better the performance of the model. Followed by XGBOOST and BPNN which performed better for crops coconut, arecanut and tapioca, banana respectively. By table 2, MAPE values for XGBOOST is quite low than any other models implemented. According to, MAPE values XGBOOST performed well for most of the crops except for tapioca; random forest performed better for tapioca with 0.226 MAPE value. By table 3, we notice that BPNN has less MAE value for all the crops, which states that BPNN performed better than any other model. Followed by ANN with MAE values ranging from 0.072 to 0.152.

CONCLUSION

In this research work, we worked on four different crops – Tapioca, Coconut, Banana, Arecanut and tried to predict the yield of all the four crops. Kerala crop data was used to predict the yield of the crops selected. We used four different models to predict the yield and evaluated each model performance using three different metrics. Upon evaluating the performance using three different metrics- RMSE, MAPE, MAEit is observed that XGBOOST is performing better compared to the other three for most of the crops i.e., for Coconut, Banana, Arecanut. Random Forest performed better for Tapioca and it also stands second after XGBOOST in yielding good results. Finally, after carefully assessing the results, we conclude that machine learning models outperformed deep learning models in this task of predicting crop yield. Deep learning methods will be giving good results if we use large number of data's. With less number of data's machine learning methods are giving better results than deep learning methods.

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Our research was limited to four crops in Kerala. Further improvements can be made to this by adding a greater number of crops and considering various states or even a different country to get a wider perspective of this research.

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Table 1. RMSE values for implemented models

CROPS	RF	XGBOOST	ANN	BPNN
TAPIOCA	0.015	0.130	0.179	0.119
COCONUT	0.037	0.070	0.168	0.086
BANANA	0.031	0.092	0.151	0.061
ARECANUT	0.015	0.052	0.097	0.062

Table 2. MAPE values for implemented models

CROPS	RF	XGBOOST	ANN	BPNN
TAPIOCA	0.226	0.681	0.562	11.011
COCONUT	0.290	0.177	1.033	0.592
BANANA	0.252	0.247	0.350	0.584
ARECANUT	0.339	0.206	0.463	0.569

Table 3. MAE values for implemented models

CROPS	RF	XGBOOST	ANN	BPNN
TAPIOCA	0.124	0.240	0.152	0.093
COCONUT	0.190	0.049	0.127	0.068
BANANA	0.252	0.115	0.117	0.051
ARECANUT	0.113	0.039	0.072	0.051

Fig 1. Work Flow

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RESEARCH ARTICLE
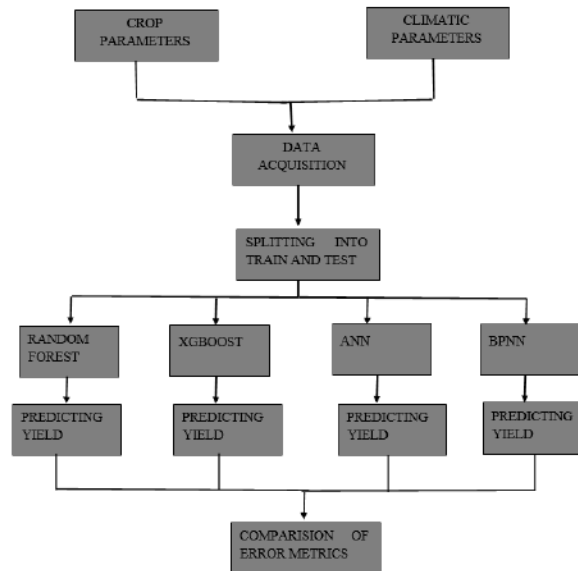
Plant Leaf Disease Image Resolution Classification using Deep Convolution Neural Network

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ABSTRACT

In agricultural operations, one of the main processes is to effectively identify and classify the crop leaf diseases. In the past decades, many deep learning models have been applied to feasibly and efficiently detect and classify the crop leaf diseases. Among many models, a Dual-Attention and Topology-Fusion with Generative Adversarial Network (DATFGAN) has achieved better accuracy to categorize the crop leaf diseases based on the texture features. On the other hand, the GAN aims at training a generator that models a mapping from a prior latent distribution to the real data distribution. The DATFGAN training could be accelerated highly by developing improved algorithm to coordinate generator and discriminator. Thus, it is crucial to learn the spatial relationships across a series of observations. Thereforein this article, Positional-aware DATFGAN (PDATFGAN) model is proposed to learn a coordinate manifold that is orthogonal to the latent distribution manifold. In this model, a Positional-aware GAN (PGAN) is introduced in which the generator creates images by parts according to their spatial coordinates as the condition. Once a latent vector is sampled, the generator conditions on every spatial coordinate and creates patches at every resultant spatial location. Also, the discriminator learns to decide whether neighboring patches are homogeneous and continuous across the edges between many patches. After that, the created high-resolution image patches are combined to get the full leaf image.

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Further, the leaf images are fed to the Deep Convolutional Neural Network (DCNN) classifier for classifying the crop leaf diseases. So, conditional coordination in DATFGAN can able to generate high-quality images than the quality of DATFGAN only. This enables the low-quality image leaf disease classification more robust. Using the generation by parts property, the PDATFGAN is greatly parallelable and intrinsically inherits the standard divide-and-conquer design paradigm which allows large field-of-view image generation. Finally, the experimental results reveal that the PDATFGAN outperform the state-of-the-art deep learning models.

Keywords: Crop leaf disease, Deep learning, DATFGAN, Conditional coordinates, Spatial correlation, CNN

INTRODUCTION

Leaves play a crucial role in crop production for providing data about the quantity and quality of crop yield. Various factors influence food production including global warming, weeds and soil erosion. Further, the development of a number of agricultural products and a source of economic losses pose a worldwide challenge to plant and leaf diseases [1]. The inadequacy of the usage of pesticide/fungicide results in a lack of identifying the infections/bacteria/virus in plants. So, plant leaf diseases have been commonly taken into account in the research field with an emphasis on the genetic characteristics of diseases. Precision agriculture incorporates the most sophisticated technologies for decision-making optimization. The visual analysis and biomedical examinations are typically performed by diagnosing plants when needed. But, this approach normally takes time and is economically unsuccessful. To solve these problems, advanced and intelligent techniques are required to identify the plant leaf diseases. The standard machine learning algorithms have been used in several researches to conduct the agricultural activities [2]. Nowadays, deep learning as a subcategory of machine learning has been remarkably successful in the identification and classification of real-life objects. As a result, an agricultural research has been progressed towards the deep learning-based solutions [3]. The deep learning techniques have been realized classical results to execute the farming activities such as crop discrimination [4], fruit harvesting [5] and crop detection [6]. Likewise, current researches have also concentrated on the other significant farming problem of crop/plant leaf disease detection [7].

Many standard deep learning algorithms have been applied to classify the plant leaf diseases through well-known deep learning structures [8]. Also, few studies introduced modified versions of deep learning structures [9] to enhance the efficiency of classifying the leaf diseases in various plant species. Different DCNN and deep learning optimizers have been designed for achieving better results of leaf disease classification [10]. To identify and classify the leaf diseases, many deep learning structures include Alex Net, VGG-16, Inception-v3, Residual Network (Res Net), Google Net, and Mobile Net have been applied. The comparative study of these models has been conducted [11] to explain the impact of the fine-tuning structure by evaluating classical deep learning algorithms for leaf disease classification [12]. However; the images acquired from farms were normally unclear. The detection accuracy of pre-trained classifies were degraded due to the poor image quality. So, the low-resolution images should be super-resolved for increasing the spatial resolution and recreating the high-frequency information of sharp edges. In this regard, Dai et al. [13] introduced a DATFGAN for recognizing the crop leaf diseases. Primarily, the

unclear images were transformed into the clear and high-resolution images. Afterwards, the weight sharing scheme was used to decrease the amount of parameters and training deeper structures that categorizes the leaf diseases in relation to the texture characteristics. But, the GAN aims at training a generator [14] that models a mapping from a prior latent distribution to the real data distribution. Also, its training could be accelerated highly by devising better algorithms to coordinate generator and discriminator. Either part of the leaf or entire leaf was affected by the diseases. So, it is essential to learn the spatial relationships across a series of observations.

Hence in this article, PDATFGAN model is proposed to generate high-quality images by parts and learn the spatial relationships across a series of observations. In this model, a PGAN is introduced in which the generator creates images by parts according to their spatial coordinates as the condition. The major goal of this PGAN is to learn a coordinate manifold that is orthogonal to the latent distribution manifold. Once a latent vector is sampled, the generator conditions on every spatial coordinate and creates patches at every resultant spatial location. Also, the discriminator learns to decide whether neighboring patches are homogeneous and continuous across the edges between many patches. Thus, conditional coordination in DATFGAN has the probable of creating high-quality images than the quality of DATFGAN alone. This enables the low-quality image leaf disease classification more robust. Using the generation by parts property, the PDATFGAN is greatly parallelable and intrinsically inherits the standard divide-and-conquer design paradigm which allows large field-of-view image generation. The rest of the article is prepared as follows: Section II studies the existing studies related to the plant leaf diseases using deep learning models. Section III describes the methodology of PDATFGAN and Section IV illustrates its performance efficiency. Section V summarizes the research work and provides the future scope.

LITERATURE SURVEY

Fuentes et al. [14] suggested a robust deep learning-based detector to recognize the real-time tomato plant diseases and pests. In this model, Faster Region-based CNN (FR-CNN), Region-based Fully Convolutional Network (R-FCN) and Single Shot Multibox Detector (SSD) were combined with deep feature extractors, namely VGG net and Res Net. Also, a method was proposed for local and global class annotation as well as data augmentation for increasing the accuracy and reducing the number of false positives during training. But, few class labels with high pattern variation tend to be confused with others due to the inadequate number of images, resulting in high false positives. Singh et al. [15] developed a Multilayer CNN (MCNN) for classifying the mango leaves infected by the Anthracnose fungal disease. First, the healthy and infected leave images were collected for mango tree suffering from fungal disease. Then, the images were pre-processed using histogram of equalization that balance the invariability among images captured in real conditions. These images were resized to a typical size image by the central square crop scheme. After, the MCNN-based ternary classifier was trained and tested for detecting the diseased mango leaves. But, its compatibility to classify multiple diseases was not effective due to the use of softmax function.

Sibiya & Sumbwanyambe [16] designed a Neuroph Studio framework as an IDE for constructing the highly facilitated DCNN wherein the convolution and pooling feature extractions were embedded in the Neuroph library. The CNN was constructed with 50 hidden layers to recognize and classify the maize leaf diseases. Also, a back propagation was used as the learning algorithm to train the CNN model. But, it considers RGB images only whereas it needs to analyze the CNN's efficiency when considering the

gray-scale images. Nagasubramanian et al. [17] applied a new 3D-DCNN that directly assimilates the hyperspectral image. First, RGB wavebands of the hyperspectral image were used to segment the charcoal rot stem in the hyperspectral image. Then, the RGB images were transformed to HSV color space followed by segmenting the charcoal rot stem using simple thresholding. Moreover, 3D-DCNN was used for extracting the features jointly across the spatial and spectral dimension for classifying the diseased soybean stem tissue. Nonetheless, its classification accuracy was not effective. Pham et al. [18] designed a multi-class mango leaf disease classification using Deep Neural Network (DNN). At first, a wrapper-based feature selection method was performed using an Adaptive Particle-Grey Wolf metaheuristic (APGWO) for choosing the most relevant features. Then, these features were given to the Multi-Layer Perceptron (MLP) as inputs to classify the diseased leaves out of healthy leaves. However, it needs to fine-tune the MLP parameters: the number of layers, the number of hidden nodes and the activation function.

Lv et al. [19] developed a maize leaf feature enhancement method to augment the features of maize under the complex conditions. Then, a new neural network based on backbone Alexnet structure called DMS-Robust Alexnet for identifying the maize leaf diseases. In this model, dilated convolution and multi-scale convolution were combined for improving the ability of feature extraction. Batch normalization was executed to avoid over fitting when improving the robustness of the model. PReLu activation function andAdabound optimizer were used for enhancing the convergence and accuracy. But, it needs to classify more categories of maize pests and diseases simultaneously.

Liu et al. [20] developed a new framework called Leaf GAN for creating the images of different grape leaf diseases to train the recognition models. Initially, a generator with degressive channels was designed for creating the grape leaf disease images. Then, the dense connectivity method and instance normalization were combined into an efficient discriminator for detecting real and fake disease images through using their better feature extraction ability on grape leaf lesions. At last, the deep regret gradient penalty scheme was used for stabilizing the training process. But, training process requires different categories of images for effective identification. Yu et al. [21] suggested a novel technique of identifying apple leaf diseases using Region-of-Interest (ROI)-aware DCNN. Initially, two sub networks were designed: one was for the splitting of the input image into three different ROIs such as background, leaf region and spot region indicating the leaf diseases. The other was for the classification of leaf diseases. These two sub networks show the structure of an encoder-decoder network and VGG network, accordingly. Then, they were trained independently using transfer learning with a new training set containing class data based on the categories of leaf diseases and the ground truth images where the ROIs were removed. After, the predicted ROI feature map was stacked on the top of the input image via a fusion layer for connecting and training these sub netwroks. But, it has less classification accuracy.

Hasan *et al*. [22] recommended CNN for identifying and classifying the grape leaf diseases. In this system, three phases were performed: data input [23], feature learning and classification. First, image preprocessing was performed for enhancing the image consistency in the dataset [24] for the CNN classifier to be processed. Then, these images were fed to the CNN for training and classifying the grape leaf diseases [25]. However, the accuracy was less.

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PROPOSED METHODOLOGY

In this section, the PDATFGAN model with DCNN for identifying and classifying the crop leaves diseases is briefly explained.

Positional-Aware GAN

The classical GAN generates a full image automatically and so image resolutions are limited by memory or computational restraints. To avoid this issue, the Positional-aware GAN (PGAN) is proposed in this study which produces a certain patch of an image conditioned on a spatial location instead of the full image at a time. The produced images are after fused together to create a globally coherent absolute image. The PGAN comprises two networks: a generator (G) and a discriminator (D). The generator encompasses two coordinate systems with shallow-feature extraction network: a fine-grained micro coordinate system for G and a coarse-grained macro coordinate system for D and images of 3 dimensions: entire images (actual (a) and generated (x)), macro patches (actual $(a^{'})$ and generated $(x^{'})$) and micro patches (generated $(x^{''})$). Also, it includes a parameter-sharing attention-enhanced topology-fusion network and reconstruction network. The shallow-feature extraction network with a coordinate system is used to extract the shallow features and spatial location of the image patch from the generator network. Figure 1 portrays the generator network of PDATFGAN.

First, low-resolution image patches are given as inputs to the generator network and split into 2 subsets. One subset is given to an upscaling unit after the primary convolutional layer in the generator network. The other subset is given to the topology fusion network for predicting information following the second convolutional layer. Then, the Reconstruction Network (Recon Net) uses the global residual learning and fuses the upscaled image patches with predicted information for creating high-resolution image patches. Finally, the Conditional Batch Normalization (CBN) is applied to normalize the feature vectors of leaf image patches. The generator of this PGAN is a conditional model that creates high-resolution micro patches with $x^{''} = G(z, c^{''})$ where z denotes the latent vector and $c^{''}$ denotes a micro coordinate condition designating the spatial position of $x^{''}$ to be created. The ultimate target of G is to create realistic and seamless entire images through collecting a group of $x^{''}$ completely with a fusion factor φ. Basically, notice that configuring φ as a concatenation factor with no overlapping is adequate for PGAN to synthesize high-quality image patches. Observe that the dimension of the micro patches and φ also involve a cropping conversion ψ, cropping out a macro patch $a^{'}$ from an actual image a which is applied to sample actual macro patches for training D. Figure 2 portrays the discriminator network of PDATFGAN

In this configuration, the ridges between successive patches become the key problem of entire image recognition. To solve this problem, the discriminator is trained with larger macro patches which are gathered with multiple micro patches. The purpose of this model is to ensure the stability and consistency of multiple successive or neighboring micro patches in the regard of adversarial loss. The generator must reduce the gap at the boundaries between the created patches for manipulating the discriminator. This PGAN is learned with four loss factors: adversarial loss (L_{PGAN}), total perceptual loss $(L_{Content})$, spatial consistency loss (L_S) and gradient penalty loss (L_{GP}). For L_{PGAN} and L_{GP}, this PGAN collaborates only with macro and micro patches whereas classical GAN utilizes entire images a for both G and D training. Similarly, L_S is an auxiliary classifier-like GAN loss factor. Based on the configuration of φ, macro coordinate $c^{'}$ is computed for the macro patches $a^{'}$. Also, L_S and $L_{Content}$ intends to reduce the

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gap loss between the actual macro coordinate $c^{'}$ and the discriminator-approximated macro coordinate $\hat{c}^{'}$. The loss factors of PGAN are as follows:

$$\begin{cases} L_{PGAN} + L_{Content} + \lambda L_{GP} + \alpha L_S, & for D \\ -L_{PGAN} + \alpha L_S, & for G \end{cases} \tag{1}$$

Spatial Coordinate System

It is initiated with designing 2 spatial coordinate systems: a micro coordinate system for G and a macro coordinate system for D. Based on φ, every macro coordinate $c^{'}_{(i,j)}$ is related to the matrix of micro coordinates: $c^{''}_{(i,j)} = \left[c^{'}_{(i:i+N,j:j+M)} \right]$ whose absolute form is as follows:

$$c^{''}_{(i,j)} = \begin{bmatrix} c^{''}_{(i,j)} c^{''}_{(i,j+1)} \cdots c^{''}_{(i,j+M-1)} \\ c^{''}_{(i+1,j)} c^{''}_{(i+1,j+1)} \cdots c^{''}_{(i+1,j+M-1)} \\ \vdots \vdots \ddots \vdots \\ c^{''}_{(i+N-1,j)} c^{''}_{(i+N-1,j+1)} \cdots c^{''}_{(i+N-1,j+M-1)} \end{bmatrix} \tag{2}$$

During PGAN training, each pair of $c^{''}_{(i,j)}$ is equally sampled. The G conditions on every micro coordinate $c^{''}_{(i,j)}$ and learns to generate high-resolution micro patches $x^{''}_{(i,j)}$ by $G(z, c^{''}_{(i,j)})$. The matrix of created micro patches $X^{''}_{(i,j)} = G(z, C^{''}_{(i,j)})$ is generated separately when distributing similar latent vector z across the micro coordinate matrix. The design concept of generating $c^{''}_{(i,j)}$ is that the created micro patches $X^{''}_{(i,j)}$ must be spatially nearer to every other. After, the micro patches are fused by φ to produce an absolute macro patch $x^{'}_{(i,j)} = \varphi(X^{''}_{(i,j)})$ as a coarser partial-view of the imagery complete-scene. At the same time, $x^{'}_{(i,j)}$ is designed with a new macro coordinate $c^{'}_{(i,j)}$ under the macro coordinate system regarding $C^{''}_{(i,j)}$. On the real-time image scenario, macro coordinates $c^{'}_{(i,j)}$ are directly sampled and real macro patches $a^{'}_{(i,j)} = \psi(a, c^{''}_{(i,j)})$ are generated with ψ. Also, observe that the design selection of the micro coordinates $C^{''}_{(i,j)}$ is correlated with the topological feature of the micro/macro coordinate systems.

Moreover, a corresponding spatial coordinate matrix $C^{''}_{entire}$ is obtained based on the micro coordinate system design. This matrix is utilized for generating every micro patch needed to form an entire image separately. The created multiple high-resolution micro patches are finally combined together to form an entire leaf image.

Loss Functions

In this model, the adversarial loss L_{PGAN} allows D to distinguish between the actual macro patches $a^{'}$ and forged macro patches $x^{'}$. Also, it supports G to deceive D with seemingly realistic micro patches $x^{''}$. It is defined as:

$$L_{PGAN} = \mathbb{E}_{a,c^{'}}\left[D\left(\psi(a, c^{'}) \right) \right] - \mathbb{E}_{z,C^{''}}\left[D\left(\varphi\left(G(z, C^{''}) \right) \right) \right] \tag{3}$$

In Eq. (3), $c^{'}$ and $c^{''}$ are coordinates for macro patches on D and micro patches on G. Observe that $G(z, C^{''})$ is the micro patches are produced by separate tasks. Also, gradient penalty is applied to the macro patches differentiation:

$$L\_GP = \mathbb{E}_{\dashv}(x\hat{}\,{}^{'})\mathbb{[}(\|\nabla\_(x\hat{}\,{}^{'})D(x\hat{}\,{}^{'})\|\_2 - 1)^2] \tag{4}$$

In Eq. (4), $x\hat{}^{'} = \varepsilon x\hat{}^{'} + (1-\varepsilon)x\hat{}^{'}$ is computed between randomly coupled $x\hat{}^{'}$ and $a\hat{}^{'}$ with a random number $\varepsilon \in [0,1]$. At last, the spatial uniformity loss L_S is equal to auxiliary classifier-like GAN loss. The discriminator is equipped with an auxiliary prediction head A which intends to measure the macro coordinate of a given macro patch with $A(a\hat{}^{'})$. A minor variation is that both $c^{''}$ and $c^{'}$ have comparatively more continuous ranges than the discrete configuration of auxiliary classifier-like GAN. So, a gap measurement loss is applied for L_S. It intends to learn G for producing equivalent micro patches by $G(z, c\hat{}^{''})$ regarding the given spatial condition $c\hat{}^{''}$. The spatial uniformity loss is defined as:

$$L\_S = \mathbb{E}_{\top}(c\hat{}^{'})^{\boxed{i0}} \llbracket \|c\hat{}^{'} - A(a\hat{}^{'})\|\_2 \rrbracket \qquad (5)$$

Conditional Batch Normalization
This positional-aware GAN utilizes CBN to normalize and modulate the features by conditionally generate γ and β which used in standard batch normalization. It determines $o\_K = (((i\_K - \mu\_K))/\sigma\_K) * \gamma + \beta$ for the $K\hat{}th$ input feature i_K, output feature o_K, feature mean μ_K and feature variance σ_K. But, positional-aware GAN gives both spatial coordinate and latent vector as conditional inputs which both containing real values rather than normal discrete classes. So, 2 MLPs: $\llbracket MLP \rrbracket\_\gamma(z,c)$ and $\llbracket MLP \rrbracket\_\beta(z,c)$ are created for every CBN layer that conditionally generates γ and β.

DCNN-based Crop Leaf Diseases Classification
After creating the complete high-resolution plant leaf images, DenseNet-121, Mobile Net V2 and Shuffle Net V2 structure-based DCNN classifiers are selected as classification networks. During the training stage of these classifiers, most of the weights in the actual models are maintained and only the softmax layers are trained. Adam is utilized as an optimizer and cross-entropy is used as a loss factor. Table 1 displays the training details of these models. Finally, these trained classifiers are used to classify the test leaf images into 15 different categories: bell pepper bacterial spot, bell pepper healthy, potato early blight, potato healthy, potato late blight, tomato target spot, tomato mosaic virus, tomato yellow leaf curl virus, tomato bacterial spot, tomato early blight, tomato healthy, tomato late blight, tomato leaf mold, tomato septoria leaf spot and tomato two spotted spider mite.

EXPERIMENTAL RESULTS
In this section, the performance of PDATFGAN is analyzed by implementing it in Python 3.7.8 using the plant leaf images with their corresponding labels. These images are collected from Plant Village Dataset (PVD) at https://www.kaggle.com/emmarex/plantdisease.This dataset comprises 20636 images of diseased and healthy plant leaves of tomato, bell-pepper and potato. In this analysis, totally 2250 images are considered. For training, 1500 images are considered in this analysis i.e., 100 images in each class are selected randomly whereas 750 images are considered for testing i.e., 50 images in each class. Also, its efficiency is compared with DATFGAN in terms of precision, recall, f-measure and accuracy to know its improvement on classification of crop leaf diseases.

Precision
It refers to the number of correctly classified plant leaf diseases i.e., diseased leaves at True Positive (TP) and False Positive (FP) rates.

$$Precision = (No.\,of\,correctly\,classified\,diseased\,leaves)/(No.\,of\,correctly\,classified\,diseased\,leaves + No.\,of\,incorrectly\,classified\,diseased\,leaves)$$

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Figure 3 depicts the precision of different classifier models for classifying leaf diseases using raw images and images restored by DATFGAN and PDATFGAN. It analyzes that the precision of MobileNet V2 for both raw and restored leaf images is higher than the other classifier structures such as ShuffleNet V2 and DenseNet-121. For instance, the precision of PDATFGAN with MobileNet V2 is 1.48% higher than the ShuffleNet V2 and 0.14% higher than the DenseNet-121 classifiers.This is achieved because of extracting spatial coordinates of leaf images along with the shallow features for classification of different types of plant leaf diseases (classes).

F-measure

It is the harmonic average of precision and recall.

$$F - measure = 2 \times (Precision \cdot Recall)/(Precision + Recall) \qquad (8)$$

Figure 4 depicts the f-measure of different classifier models for classifying leaf diseases using raw images and images restored by DATFGAN and PDATFGAN. It analyzes that the f-measure of MobileNet V2 for both raw and restored leaf images is greater than the ShuffleNet V2 and DenseNet-121. For instance, the f-measure of PDATFGAN with MobileNet V2 is 1.48% higher than the ShuffleNet V2 and 0.14% higher than the DenseNet-121 classifiers. So, it addresses that the MobileNet V2 with PDATFGAN model achieves better f-measure compared to the other classification models.

Accuracy

It is the ratio of exact classification of plant leaf diseases over the total amount of attempts executed.

$$Accuracy = (TP + True\ Negative\ (TN))/(TP + TN + FP + FN) \qquad (9)$$

TP is an outcome that classifier exactly classifies the diseased leaves as itself. TN is an outcome that classifier exactly classifies the healthy leaves as itself. FP is an outcome that classifier inexactly classifies the diseased leaves as healthy. FN is an outcome that classifier inexactly classifies the healthy leaves as diseased leaves. Figure 5 depicts the overall classification accuracy (%) of different classifier models for classifying leaf diseases using raw images and images restored by DATFGAN and PDATFGAN. It notices that the accuracy of Mobile Net V2 for both raw and restored leaf images is increased than the Shuffle Net V2 and DenseNet-121. For example, the accuracy of PDATFGAN with Mobile Net V2 is 1.48% increased than the Shuffle Net V2 and 0.14% increased than the DenseNet-121 classifiers. This is attained due to the transformation of low-resolution images into the high-resolution images at patch-level without consideration of the entire leaf images for leaf diseases classification.

CONCLUSION

In this study, a PDATFGAN model is proposed to classify the types of plant leaf diseases. First, different plant leaves images of diseased and healthy conditions are acquired as training and testing set. Then, training images are given to the PDATGAN model which uses spatial coordinates as a condition to learn the spatial correlation across a series of observations. In this model, the generator creates high-quality image patches at each spatial position based on the conditions on each spatial coordinates. The discriminator learns to confirm whether neighboring patches are homogeneous and continuous across

the edges between many patches. After that, the created high-resolution image patches are combined to get the full leaf image. Further, DCNN-based classifier is applied to categorize the crop leaf diseases. Finally, the experimental results proved that the PDATFGAN with Shuffle Net V2, DenseNet-121 and Mobile Net V2 classifier achieves an overall classification accuracy of 90.32%, 91.54% and 91.86%, respectively whereas the DATFGAN with Shuffle Net V2, DenseNet-121 and Mobile Net V2 classifier achieves an overall classification accuracy of 90.88%, 91.52% and 91.69%, respectively. Therefore, this concludes that the Mobile Net V2 classifier with PDATGAN model achieves a high accuracy than all other classifiers with DATGAN model.

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Table 1. Training Parameters

Model	Learning rate	Batch size	Epochs
DenseNet-121	0.0005	10	50
MobileNet V2	0.0001	10	60
ShuffleNet V2	0.0001	10	70

Figure 1. Structure of Generator Network	Figure 2. Structure of Discriminator Network
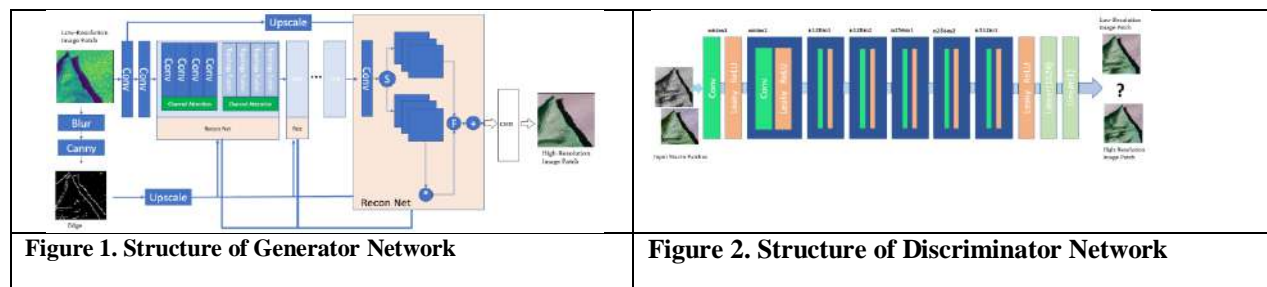
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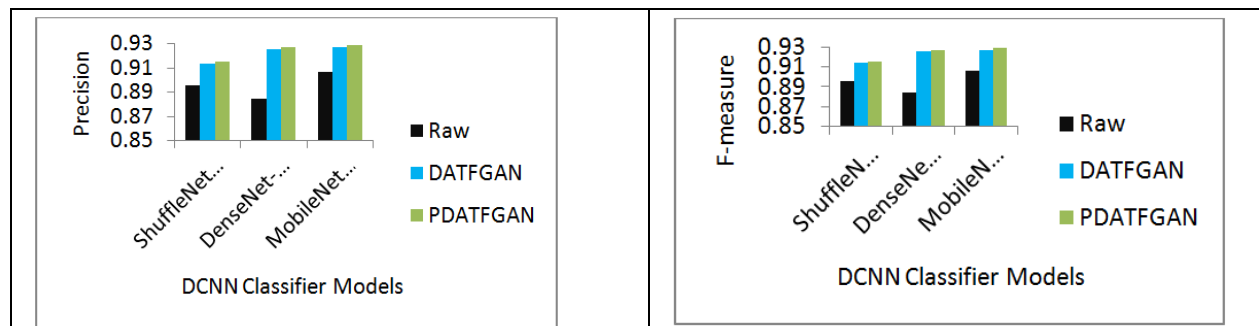
Figure 3. Comparison of Precision	Figure 4. Comparison of F-measure
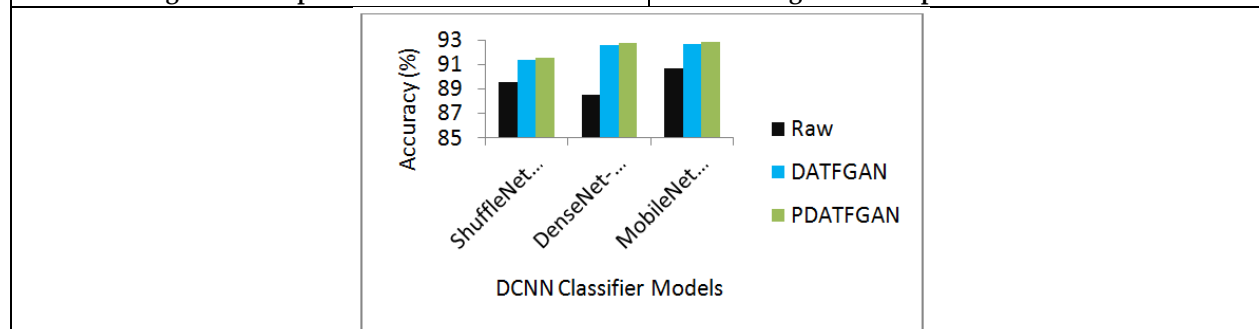
Figure 5. Comparison of Accuracy

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RESEARCH ARTICLE

Optimal Cloud Load Balancing Approach using Dynamic Programming Techniques

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ABSTRACT

Cloud computing in the discipline of computer science plays a essential position in statistics and carrier get admission to for the customers if you want to show off the centers in phrases of software, hardware and computation as offerings. The monstrous reputation and huge getting access to nature growth in exponential fashion due to its restriction-unfastened on licenses, speedy updating and smooth to deal with right care. Nowadays popular agencies like Oracle, Google, IBM, Microsoft, Apple, Samsung and so forth offer cloud as a carrier to all in their customers based totally on their norms and pricing. The boom in cloud gaining access to makes the provider carriers face the number one issue in terms of load balancing. The trouble due to mistaken load balancing in cloud servers will have an effect on the entire commercial enterprise method. This paper offers the most fulfilling cloud load balancing method for effective service having access to towards cloud services for the simultaneous cloud having access to requests in variable standards's. In the close to destiny, this paper may be prolonged with artificial intelligence based totally load balancing approach for the upcoming or anticipated scale of load balancing problems.

Keywords: Cloud, Dynamic programming, Load balancing, artificial intelligence

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INTRODUCTION

Cloud Computing
Cloud computing is one of the regions of facts era currently considered an alternative to standard fashions of information processing that uses the net and significant far-flung servers to maintain statistics and applications. Such structures permit far off statistics processing, offering fulfillment of high performance that fail in both safety and availability of the statistics infrastructure [16].

Load Balancing
Load balancing is a method used to distribute workloads uniformly throughout servers or different compute assets to optimize network performance, reliability and potential. [17,18].

Dynamic Programming
Dynamic Programming is a method for solving a complex problem by breaking it down into a collection of simpler sub problems, solving each of those sub problems just once, and storing their solutions using a memory-based data structure (array, map, etc).

Literature Review

Technological development in pc global and rapid trends of disbursed systems over previous couple of years induced that load balancing problem were took into attention as a chief challenge greater than ever in those systems. This section offers a few essential ideas and techniques of load balancing mechanism. Load balancing is the technique of redistributing the general device paintings load among all nodes of the distributed system (community hyperlinks, disk drivers, central processing devices…) to improve both resource utilization and process reaction time whilst averting a state of affairs where some nodes are overloaded at the same time as others are underneath loaded or idle . Load balancing is a important and inseparable a part of cloud computing and elastic scalability. There are many numerous kinds of load balancing mechanisms and approaches which maximum of the research have been labeled as two major classes [1,2,3]: static and dynamic. In static strategies [4,5,6], there are normally prior information and some assumptions approximately the global repute of the system along with activity resource necessities, communication time, processing strength of system nodes, memory and garage devices ability and so forth.

A static technique is a kind of project from a set of duties to a fixed of assets which could take both a deterministic or a probabilistic form [2]. In addition, this method is defined usually within the design or implementation of structures [7]. In deterministic assignments, the extra workload of a certain node may be transferred to every other specific node all of the time, but in probabilistic tactics each node sends its more responsibilities with possibility P to a node and with opportunity 1-P to any other one. The important disadvantage of static load balancing algorithms is that the contemporary country of the device is not taken into consideration when making the decisions and consequently it isn't a appropriate technique in systems consisting of allotted structures which maximum states of the gadget changes dynamically.

Dynamic load balancing techniques take into consideration the modern state of structures that their decisions are based on. In this method tasks can move dynamically from an overloaded node to an beneath-loaded one and that is the principle gain of dynamic load balancing algorithms which could trade constantly in step with the present day nation of the machine. However designing and enforcing a dynamic load balancing algorithm is a good deal extra complicated and tougher than finding a static answer, but thru dynamic mechanisms we can advantage a higher overall performance and have more accurate and efficient answers [1,2]. Dynamic load balancing algorithms can be designed in two specific approaches: dispensed and non-dispensed. In allotted tactics e.g. [8,9,10,11], the load balancing procedure can be completed by using all nodes within the system.

In addition, in this technique all nodes can speak with each other for reaching a global intention within the device which is called cooperative or each node can paintings independently for simply reaching a neighborhood purpose that is non cooperative form. But, in a non-distributed scheme [12,13,14,15], the responsibility of balancing the gadget workload could not be carried out via all machine nodes. In centralized technique in non-distributed scheme; a unmarried node best can execute the load balancing mechanism amongst all nodes. In semi-dispensed shape, the gadget may be divided into a few partitions or clusters and a single node execute the burden balancing process in each partition.

PROPOSED METHODOLOGY

The following figure-4 represents the proposed methodology for optimal cloud content access load balancing approach. It includes the unification of four different sub servers for effective cloud-based content or service accessing.

They are:
- Rapid Cloud Request and Authentication Server.
- Conditional Cloud Request Domain Routing
Server.
- Computable Virtual Load Balancer.
- Countable Mirror Request Content Handler Server.

Rapid Cloud Request and Authentication Server:
The verification and validation of client requests are processed by registered username and passwords with the associated security algorithms for data security. It processes the client requests through dynamic programming by classifying the request types into applications, mails, video streaming and data retrievals[8].

Conditional Cloud Request Domain Routing Server:
The client request classifications are verified with their protocols, https for applications, SMTP and POP3 for mail transfers, UDP for video streaming and TCP for data retrievals[6].

Computable Virtual Load Balancer: The level of significance for the exact classification of client cloud requests are min 5% with inappropriate classification that can be dealt as a metadata with proper mapping for client-side accurate data accessing including the voice over internet protocol[7].

Countable Mirror Request Content Handler: More than one request for the same cloud data or service at the same time will affect the service performance which can be handled with a single high capability server with virtual duplications and multiple physical servers with the proper divergence of service towards specific content[5].

Implementation
Consider the sample cloud service as Trichy Justin cloud Services as follows:

Authentication Server
The code used for password authentication using JSON is as follows:
curl -X POST
https://identity.api.trichyjustincloud.com/v2.0/token s -d
'{"auth":{"passwordCredentials":{"username":"theUserName","password":"thePassword"}}}' -H
"Content-type: application/json"

Domain Routing Server For The Sample Image Reference

The following sample code is used for domain
routing based content services in cloud computing,

```
{
"server": {
"name": "new-server-test",
"min_count": 1,
"max_count": 1,
"imageRef": "imagebinary.ref ",
"flavorRef": "2",
"diskConfig": "auto",
"metadata": {
"My Server Name": "Ubuntu 10.04 LTS"
},
"personality": [{
"path": "/etc/image.txt",        "contents":
"9WQUICAgICAgDQoiQSBjbG91ZCBkb2VzIG5vd CBrbm93IHdoeSBp
dCBtb3ZlcyBpbiBqdHN0IHN1Y2ggYSBkaXJlY3
Rpb24gYW5k
IGF0IHN1Y2ggYSBzcNGVlZC4uLkl0IGZlZWxzIG FuIGltcHVs
c2lvbi4uLnRoaXMgaXMgdGhlIHBsYWNlIHRvI
GdvIG5vdy4g
QnV0IHRoZSBza3kga25vd3MgdGhlIHJlYXNvbn
MgYW5kIHRo
ZSBwYXR0ZXJucyBiZWhpbmQgYWxsIGNsb3Vkc ywgYW5kIHlv
dSB3aWxsIGtub3csIHRvbywgd2hlbiB5b3UgbGlmd CB5b3Vy
c2VsZiBoaWdoIGVub3VnaCB0byBzZWUgYmV5b
25kIGhvcml6b25zLiINCg0KLVJpY2hhcmQgQmFjaA=="       }]
}
}
```

The Sample server's response from the cloud serverside towards the client is as follows,

```
{
"server": {
"OS-DCF:diskConfig": "AUTO",
"adminPass": "justin2020Trichy",
"id": "image-reference-003", //Image Id      "links": [{        "href":
"https://dfw.servers.api.trichyjustincloud.com/v2/ser vers/ image-reference-003",
"rel": "self"
}, {
"href":
"https://dfw.servers.api.trichyjustincloud.com/serve rs/ image-reference-003",
"rel": "bookmark"
}]
}
}
```

Virtual Server

The virtual server can handle any type of metadata which are all the exception from the classification of client-side requests based on protocols and content variations. The responses from the cloud server are as follows,

Responses
"created": "2020-04-07T13:23:29Z",
"flavor": {
"id": "1",
"links": [{"href":
"https://dfw.servers.api.trichyjustincloud.com/flavor
s/1",
"rel": "bookmark"
}]
},
"image": {
"id": "image-refrence-id-002", "links": [{"href":
"https://dfw.servers.api.trichyjustincloud.com/imag es/ image-refrence-id-002",
"rel": "bookmark"
}]
},
"key_name": "",
"links": [{
"href":
"https://dfw.servers.api.trichyjustincloud.com/v2/20
20/",
"rel": "self"
}, {
"href":
"https://dfw.servers.api.trichyjustincloud.com/2020/
",
"rel": "bookmark"
}],
"metadata": {},
"name": "testinstance",
"progress": 100,
"status": "ACTIVE",
}
}

Multiple Servers Handling

The multiple requests for the same data accessing can be handled with different options as a single server with maximum load incorporated with multiple virtual ids or variational servers with individual physical address strategy for handling the client requests efficiently.

<? xml version="1.0" encoding="UTF-8"?>
<server xmlns:RAX-
SERVER="https://docs.justincloud.com/servers/api/ ext/server_bandwidth/"
xmlns="http://docs.openstack.org/compute/api/v1.1
" >

<image id=" image-reference-id-002"><atom: link rel="self" href="http://servers.api.trichyjustincloud.com/v2/im ages/ image-refrence-id-002"/><atom: link rel="bookmark" href="http://servers.api.trichyjustincloud.com/imag es/ 2020"/>
</image>
<flavor id=" image-reference-id-003"><atom: linkrel="self"href="http://servers.api.trichyjustincloud.com /v2/flavors/2021"/>
<atom: link rel="bookmark" href="http://servers.api.trichyjustincloud.com/flavors/2022"/></flavor>
<metadata>
<meta key="JustinCloud Label">Page Head
1</meta>
<meta key="Image Version">2.1</meta>
</metadata>

<addresses>
<network id="public">
<ip version="4" addr="192.168.1.1"/>
<ip version="6" addr="::cade:192.168.1.2"/><ip version="4" addr="192.168.1.3"/><ip version="6" addr=": cade:
8516:0B32"/>
</network>

<network id="private">
<ip version="4" addr="192.168.1.4"/>
<ip version="6" addr="::cade:192.168.1.5"/>
</network>

</addresses>

<atom: link rel="self"
href="http://servers.api.trichyjustincloud.com/v2/12
34/servers/2020"/><atom: link rel="bookmark" href="http://servers.api.trichyjustincloud.com/1234/ servers/2021"/>

</server>

RESULTS AND DISCUSSION

The sample request table by maintaining the JSON server yields the following results with the local server maintenance id named as Justin cloud (local sample server). The priority P1 is given for web content, P2 for API requests, P3 for Mail requests and P4 for streaming requests in order to balance the server by reducing the task level by finishing the lower to higher load executions as part of combining smaller to bigger solutions in dynamic programming. The following table shows the client request table for JSON sample server for the local cloud entertainment domain justincloud.com with two specific time intervals.

- After applying the authenticity and role of the client priority, cased service results are as follows:
- Now focus on to the conditional cloud request domain routing server   for client request categorization as follows:

```
public function getRequestcontent () {
//Switch        switch ($allArrayKeys [0]) {            case 'API':            $result = $resp;            break;    case 'Mail':
$result = $resp;            break;            case 'Stream':            $result = $resp;            break; case 'Web':
$result = $resp;        break;        break;
}
//dd ($result);
```

return view ('B2B: pages.contenttoshow ()', compact ('result'));
}
}

Based on the dynamic programming, the entire client requests are initially divided into two categories. They are priority-based and non-priority based in which the cloud service is effective if and only if it satisfies the prime customers with max buffers of load service followed by the available resource load balancing with proper care for non-prioritized client requests. Now the classification table is as follows:

The protocol based classification table for API accessing includes the HTTP protocol and Secured HTTPS protocol so that the Https are more reliable for prioritized client request response system for further processing is as follows:

The each individual service Ids based on priority emphasized API request with secured HTTPS data service provisioning is as follows:

The client request classification table for Mail accessing is as follows:

The each individual service Ids based on priority emphasized Mail access request with SMTP protocol first data service provisioning is as follows:

The client request classification table for Stream accessing is as follows:

The each individual service Ids based on priority emphasized streaming access request with Real-time message protocol which is the popular and easier data service provisioning as follows:

The client request classification table for basic web content accessing is as follows:

The each individual service Ids based on priority emphasized web content access request with TCP protocol which is the popular and easier data service provisioning as follows:

Now the priority based Web content accessing rights with data and service responses are provided in the following order as in the below table.

The order of service completion based on the in the following diagram

The workflow for load balancing table is represented dynamic programming table is as follows:

The following table illustrates the efficiency achieved with the proposed model for dynamic programming implementation in cloud server service load balancing schema is as follows:

The following diagram shows the efficiency of the proposed load balancing schema for dynamic programming based Cloud service schema results.

The proposed methodology yields the effective load balancing which will enforce the cloud service with the maximum level of satisfaction. The final result provides an improvement efficiency of 59.5%.

CONCLUSION

Load balancing in cloud computing service requires a complex mechanism which includes the pleasant and amount of provider supply with the concern-based approach to improve the enterprise model with powerful attainments. This paper offers with the unification model which adopts the dynamic programming method of classifying the requests with right care on precedence and non-priority implementation on services and deal with the individual thing based on their request type and content requirement with the simultaneous reproduction request coping with at the equal time. The number one approach offers with the authentication and verification of legitimate patron based on their registered records. The successive approach specializes in client request class primarily based at the type of request alongside the protocol associated with them for proper amalgamations. The next approach acts as a digital server aspect for coping with metadata as a mixture of requests raised from the patron-facet. The final method deals with the multiple servers handling for dealing with the duplicated or more than one requests carrier issuer for simultaneous accessing. The proposed dynamic programming consequences are blended collectively which yield the load balancing performance with 59.55 improvements. In the near destiny, this paper might be extended with the implementation of artificial intelligence for the future requirement in load balancing in cloud computing server provider for the customer-facet access.

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Table-1: Sample Cloud Server Service Request Table

No of clients	Requests	Time Intervals
43	56	T1
75	112	T1
106	314	T1
62	126	T2
81	253	T2
94	149	T2

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Table-2: Priority-based Cloud Server Service Request Table

No of clients	Requests	Time Intervals	Priority queue
32	45	T1	2
56	73	T1	1
65	171	T1	3
43	67	T2	3
60	136	T2	2
57	92	T2	1

Table-3: Cloud Content Server Service Request Type Table

No of clients	Req	API	Mail	Streaming	Web	Time Intervals	Priority queue
32	45	16	8	15	6	T1	2
56	73	24	17	22	10	T1	1
65	171	52	29	72	18	T1	3
43	67	20	4	29	14	T2	3
60	136	47	9	63	17	T2	2
57	92	19	7	45	21	T2	1

Table-4: Protocol based API Cloud Server Service Request Table

Time Priority ID	API	HTTP	HTTPS*
T12	16	11	5
T11	24	17	7
T13	52	29	23
T23	20	13	7
T22	47	28	19
T21	19	11	8

Table-5: Individual Cloud Server Service Request ID Table

Client Service ID for API with Priority-2	Service Counts
P2T12hs1,P2T12hs2,P2T12hs3,P2T12hs4, P2T12hs5	5
P2T11hs1,P2T11hs2,P2T11hs3,P2T11hs4, P2T11hs5,P2T11hs6,P2T11hs7	7
P2T13hs1,P2T13hs2,P2T13hs3,P2T13hs4, P2T13hs5,P2T13hs6,P2T13hs7,P2T13hs8, P2T13hs9,P2T13hs10,P2T13hs11,P2T13hs 12, P2T13hs13,P2T13hs14,P2T13hs15, P2T13hs16,P2T13hs17,P2T13hs18,P2T13 hs19,P2T13hs20,P2T13hs21,P2T13hs22,P 2T13hs23.	23
P2T23hs1,P2T23hs2,P2T23hs3,P2T23hs4, P2T23hs5,P2T23hs6,P2T23hs7	7
P2T22hs1,P2T22hs2,P2T22hs3,P2T22hs4, P2T22hs5,P2T22hs6,P2T22hs7,P2T22hs8, P2T22hs9,P2T22hs10,P2T22hs11,P2T22hs 12,P2T22hs13,P2T22hs14,P2T22hs15,P2T 22hs16,P2T22hs17,P2T22hs18,P2T22hs19	19
P2T21hs1,P2T21hs2,P2T21hs3,P2T21hs4, P2T21hs5,P2T21hs6,P2T21hs7,P2T21hs8.	8
Total	69

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Table-6: Mail Accessing Cloud Server Service Request Table

Time Priority ID	Mail	SMTP*	POP3
T12	8	7	1
T11	17	15	2
T13	29	25	4
T23	4	3	1
T22	9	8	1
T21	7	6	1

Table-7: Individual Mail accessing Cloud Server Service Request ID Table

Client Service ID for Mail access with Priority-3	Service Counts
P3T12sp1,P3T12sp2,P3T12sp3,P3T12sp4,P3T1 2sp5, P3T12sp6, P3T12sp7	7
P3T11sp1,P3T11sp2,P3T11sp3,P3T11sp4,P3T11sp5,P3T11sp6,P3T11sp7, P3T11sp8, P3T11sp9, P3T11sp10, P3T11sp11, P3T11sp12, P3T11sp13, P3T11sp14, P3T11sp15	15
P3T13sp1,P3T13sp2,P3T13sp3,P3T13sp4, P3T13sp5,P3T13sp6,P3T13sp7,P3T13sp8, P3T13sp9,P3T13sp10,P3T13sp11,P3T13sp12, P3T13sp13,P3T13sp14,P3T13sp15, P3T13sp16,P3T13sp17,P3T13sp18,P3T13sp19,P 3T13sp20,P3T13sp21,P3T13sp22,P3T13sp23, P3T13sp24, P3T13sp25.	25
P3T23sp1,P3T23sp2,P3T23sp3	3
P3T22sp1,P3T22sp2,P3T22sp3,P3T22sp4, P3T22sp5,P3T22sp6,P3T22sp7,P3T22sp8.	8
P3T21sp1,P3T21sp2,P3T21sp3,P3T21sp4, P3T21sp5,P3T21sp6.	6
Total	64

Table-8: Stream accessing Cloud Server Service Request ID Table

Time Priority ID	Streaming	UDP*	Real Time Msg Protocol (RTMP)***	DASH (Dynamic streaming over HTTP)**
T12	15	5	6	4
T11	22	8	9	5
T13	72	21	10	41
T23	29	13	7	9
T22	63	21	12	30
T21	45	12	5	28

Table-9: Individual Streaming Cloud Server Service Request ID Table

Client Service ID for streaming access with Priority-4	Service Counts
P4T12rp1,P4T12rp2,P4T12rp3,P4T12rp4, P4T12rp5, P4T12rp6	6
P4T11rp1,P4T11rp2,P4T11rp3,P4T11rp4,P4T11 rp5,P4T11rp6,P4T11rp7,P4T11rp8,P4T11rp9	9
P4T13rp1,P4T13rp2,P4T13rp3,P4T13rp4,P4T13 rp5,P4T13rp6,P4T13rp7,P4T13rp8,P4T13rp9,P4 13rp10.	10
P4T23rp1,P4T23rp2,P4T23rp3,P4T23rp4, P4T23rp5,P4T23rp6,P4T23rp7	7
P4T22rp1,P4T22rp2,P4T22rp3,P4T22rp4, P4T22rp5,P4T22rp6,P4T22rp7,P4T22rp8, P4T22rp9,P4T22rp10,P4T22rp11,P4T22rp12.	12
P4T21rp1,P4T21rp2,P4T21rp3,P4T21rp4, P4T21rp5.	5
Total	49

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Table-10: We text accessing Cloud Server Service Request ID Table

Time Priority ID	Web Content	TCP*	No P
T12	6	4	2
T11	10	9	1
T13	18	15	3
T23	14	12	2
T22	17	16	1
T21	21	18	3

Table-11: Individual web text accessing Cloud Server Service Request ID Table

Client Service ID	Service Counts
T12t1,T12t2,T12t3,T12t4	4
T11t1,T11t2,T11t3,T11t4,T11t5,T11t6,T11t7,T11t8, T11t9	9
T13t1,T13t2,T13t3,T13t4, T13t5,T13t6,T13t7,T13t8, T13t9,T13t10,T13t11,T13t12, T13t13,T13t14,T13t15.	15
T23t1,T23t2,T23t3,T23t4, T23t5,T23t6,T23t7,T23t8, T23t9,T23t10,T23t11,T23t12	12
T22t1,T22t2,T22t3,T22t4,T22t5,T22t6,T22t7,T22t8, T22t9,T22t10,T22t11,T22t12,T22t13,T22t14,T22t15 ,T22t16.	16
T21t1,T21t2,T21t3,T21t4, T21t5,T21t6,T21t7,T21t8, T21t9,T212t10,T21t11,T21t12,T21t13,T21t14,T21t15,T21t16, T21t17, T21t18.	18
Total	74

Table-12: Individual Priority based web Cloud Server Service Request ID Table

Services-Order Based	Priority Services Count	Non Priority Services count	Multiple Servers / Content count	Actual Service Time	Proposed Dynamic service Time (Priority)	Proposed Dynamic service Time (Non Priority)
Web access with Priority P1	74	12	3	86	24.67	4
API access with Priority P2	69	109	2	178	34.5	54.5
Mail access with Priority P3	64	10	2	74	32	5
Streaming access with Priority P4	49	197	3	246	16.33	65.67
			Total	584	107.5	129.17

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Table-13: Proposed model results for Load balanced Cloud Server Service

Service Type	Service Time
Actual Cloud Service without load balancing	584
Priority Based Load Balancing	107.5
Non Priority Based Load Balancing	129.17
Gain	100-(236.67/584)*100=59.5%
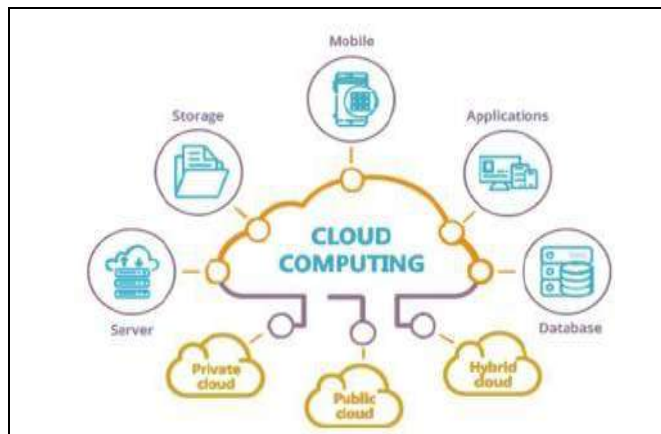
Fig-1: Cloud Computing Model
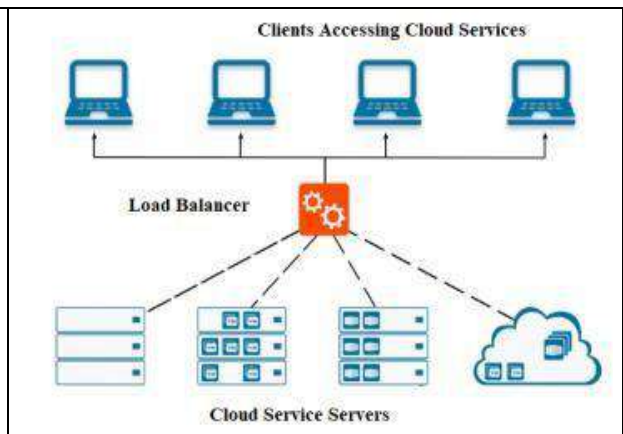
Fig-2: Load balancing role in Cloud Computing Model
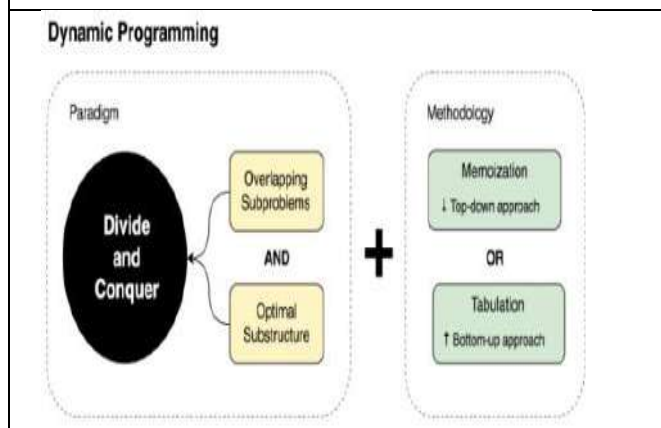
Fig-3: Dynamic Programming Approach
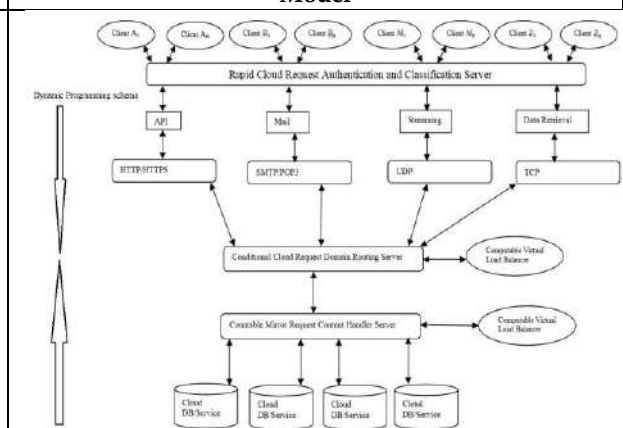
Fig-4: Proposed Load Balancing using Dynamic Programming Approach

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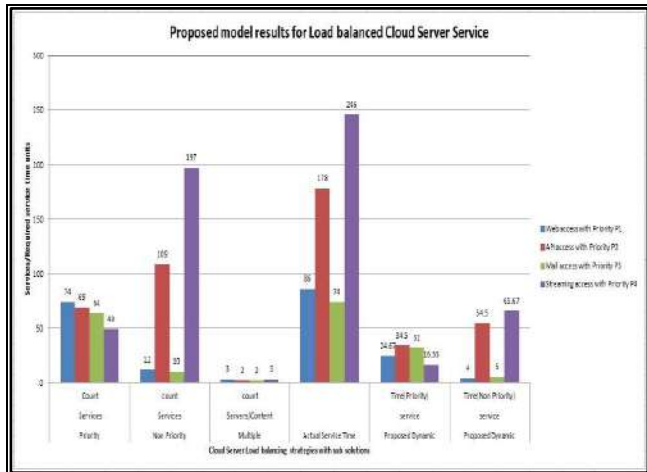
| Fig-5: Proposed Cloud Load Balancing Approach Results | Fig-6: Cloud Load balancing efficiency schema results using Dynamic Programming Approach |
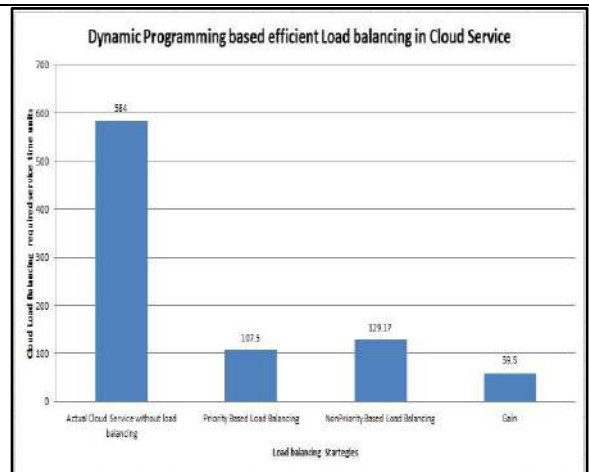
RESEARCH ARTICLE

Classification of Dementia using MRI images: A Deep Learning Approach

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ABSTRACT

Dementia is a disease found in the elderly due to damage in brain cells overtime. The damage that is caused often tends to hinder cognitive abilities in an individual. When neurons are not able to transmit signals called neurotransmitters, it can impact cognitive abilities like thinking, reasoning and memory function. Additionally, Dementia is quite often diagnosed by employing a sequence of imaging tests, associated by evaluating an individual's anamnesis. Brain imaging helped doctors perceive the depth of the disease so it could be treated efficiently. This paper contains an automated computer aided system to diagnose the depth of the disease by using a type of brain imaging test called MRI scan. This paper suggests a computer aided method to detect early dementia using MRI images. This method consists of three sub-processes: First Pre-processing, Segmentation then classification. Noise removal, Skull Stripping and Edge enhancement is applied as the pre-processing methods. The pre-processed MRI images are classified based on 4 diagnosis groups:MD, ND, Mod-D, VMD. In our paper classification is done using Inception v3 CNN architecture. And segmentation of the WM (White matter) region is done. The suggested dementia detection obtains an average classification accuracy rate of 92.26 %, with a loss of 0.2117, precision of 97.6%, F1-Score of 92.30 percent, and recall of 95.5 percent.

Keywords: Brain MRI, deep learning, CNN, segmentation, dementia, Inception v3

INTRODUCTION

Medical issues are increasing in number. Experts and numerous organizations are trying to achieve a conclusion for the diagnosis and detection of the issues of their patients. Among these, one includes the Dementia which is growing numerously in the elderly. Dementia is a term that expresses cognitive impairment and leads to symptoms related to

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loss in thinking ability, attention, memory, and logical reasoning. Dementia is quite often not a disease but is a collection of symptoms that leads to mild to severe cognitive impairment in daily life. The quick detection of dementia using MRI images would supply a threshold to treat these symptoms in no time. Dementia exists in many forms it incorporates many diseases one of them is vascular dementia. Our use case here is classification of demented brain MRI images of the Alzheimer's disease. CAD/CAMs are being implemented in order to detect different conditions in imaging of medical data and changing accuracy of diagnosis by using state-of-the-art pattern recognition as well as image processing. [1]. This condition commonly affects people aged 65 and over with only 10% of cases transpiring in young adults. That is why we diagnose this early. The changes in brain cells caused by this disease is not irreversible although it can be paused by medication provided by a verified neurologist. Classifying images is a very essential step in recognizing the pattern and getting insights of images. Earlier. Deep learning has been used in many types of image classification. Mainly Convolutional neural network has the best results for this image classification. [2] First in the section, we present the Kaggle dataset which was attained through medical image acquisition and contains both healthy and diseased brains. Signal or Image de-noising is an essential task when working with MRI images. All MRI images have a range of noises. The pre-processing is done using data augmentation, noise removal and edge enhancement [3]. For noise removal, the bilateral filter is being employed, and as for edge enhancement the histogram equalization method is implemented. Finally, the result of learning classification model and performance of the system under consideration are calculated. This includes values of Recall, Precision, F1-score, Accuracy etc.

LITERATURE REVIEW

Hierarchical extreme learning machine turned in 2020i [4], through Huizu Guietial for computer-Aided Dementia analysis. Using multilayer feature representation. DCADx, a CADx framework primarily based ionH- LM, is provided. Due to the fact CSP and BFN were proven to have higher redundancy effects on brain facts, the DCADx incorporates exclusive information red undancy discount techniques (1) CSP-primarily based DCAD(thesis,C Affemodel) indian(2)BFN –based totally DCADx. On Alzheimer's disease, the DCADx-CSP model scored 83.2 percentage. In 2019 [5], Fubao Zhu et al . proposed a multiclass deepL used, precision, accuracy, let us not forget Recall, and F1-ratingi to assess every diagnostic version in the check set. This model of DNN suggests better balance and got the fine accuracy ofi 0.88%. In 2021, Suriya Murugan et al [6]. proposed deep gaining knowledge of version for early analysis of alzheimer illnesses and dementia From MRI images. The Convolution Neural network (CNN) is used to assemble a framework for detecting particular Alzheimer's ailment symptoms from MRI snap shots. To come across dementia stages thru MRI, a Dementia network (DEMNET) has been proposed. The DEMNET has a ninety three 93% accuracy.

In 2016, slice optimization was proposed by Yin Zhang et al [7]. OASIS dataset was usedand evolved a version which shows the diseases by combining Wavelet Entropy (WE), Multi-Layer Perceptron (MLP) and Biogeography Based Optimization (BBO). By using this approach, an accuracy of 92.40% was achieved. The statistical results of their method obtained an accuracy of 92.40 ± 0.83percent, a specificity of 92.47 ± 1.23 % and a sensitivity of 92.14 ± 4.39%,.In 2018, Gang Li et al [8] used Longitudinal analysis for Alzheimer's disease diagnosis and used RNN with the OASIS Dataset. RNNis a type of looping network is used to detect the diseases of Alzheimer's. The parts of the data given is vectorized. The layer is further used to extract temporal columns thatgive details about the comparisons that have been made. In these two layers there exists a binary layer for classes and fully connected layer. Training and testing and then validating the accuracy was found to be eighty six.89% but the accuracy of MLP+BGRU is found to be 89.69 percent. In 2018, Riashat et al [9].

Implemented a Deep Convolutional Neural Networks
For Alzheimer's Disease and detection of demented mages using 3D slices. In CNN, the 3D data is pre-processed such that 2D input data is obtained. After training and testing the model using the proposed approach, an accuracy of 80.5% was achieved. This paper has an alternative approach that is fast, cheap and more reliable. Convolution Neural Networks inspired Multilayer perceptron specifically capable of image

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processing. In 2020, Zhao Fan et al [11]. hired an powerful type of MRI snap shots of Alzheimer's ailment with the aid of making use of the ADNI dataset. On this observation a characteristic class version for Alzheimer's disorder primarily based on KPCA algorithm and AdaBoost algorithm changed into constructed, and decided on 21 patients with Alzheimer's disease (advert). those outcomes display that the KPCA set of rules is getting used in the article to acquire the very best class accuracy of the 2 groups: 94.seventy seven%, the single function distinguishing capability is the node level, and the accuracy of 90.ninety four% can be performed in the imaging analysis of AD. So, in this paper we are using noise removal, data augmentation and edge enhancement for data pre-processing. For noise removal we are using Bilateral Filter and for edge enhancement Histogram Equalization (HE). White matter slicing is done using segmentation. We referred to a paper Convolution neural networks formulti- class brain ailment identification by MRI.In thisAlexNet, Vgg-16, ResNet-18, ResNet-34, ResNet-50 and Inception v3 models are used. That said, comparing their classification performance with pre-trained models are done the accuracy of 92.15% was got. We have used Inception v3 Architecture for the classification of images from the Kaggle dataset. The ideal image size for Inception v3 model is '224 X 224' pixels. The processes are highlighted in the further sections of this article. Before the images were loaded for pre-processing, we converted the image resolution of the training and testing set to 224 x 224 pixels.

METHODOLOGY

In our project we introduced the method of deep learning model as shown in figure 1. The step are: (2) Segmentation, (3) Image classification. This paper proposes an efficient method to establish and diagnose one of the most prevailing brain diseases, Dementia. We have made a diagram for the method we want to implement in Figure 1. First, we start by acquiring the dataset from Kaggle. A sample of the dataset is shown in figure 2. This data is mapped in a table format which is shown in table 1. We have reshaped our images to the size 224x224 before image processing. The processes are highlighted in the further sections of this article. Before the images were loaded for pre-processing, we converted the image resolution of the training and testing set to 224 x 224 pixels.

Data pre-processing

Images we acquired first undergo image pre-processing. Noise removal and Edge enhancement. First Data Augmentation was done as there was a class imbalance. Noise removal for test and train images is done using: The bilateral filter, this filter has proven its efficacy in MRI images in the past and thus the bilateral filter is our first approach. After this edge enhancement is done using Histogram Equalization.

Data Augmentation

Data augmentation is a pre-processing technique used to handle class imbalance and to establish a balanced dataset in each of its classes. In this article, we have implemented data augmentation to balance out two classes: Mild and Very Mild. Before augmentation it had 717 and 52 images in the train dataset respectively for each of the classes. This is depicted in table 2 with values of specific classes of the test and train dataset.

Noise Removal

We have implemented the bilateral filter for noise removal. It is called the edge preserving filter which is non-linear and it utilizes the Gaussian. Convolutional phase. The bilateral filter is given by:

$$I^{\text{filtered}}(x) = \frac{1}{W_p} \sum_{x_i \in \Omega} I(x_i) f_r(\|I(x_i) - I(x)\|) g_s(\|x_i - x\|),$$

Here, the normalization term Wp is represented by,

$$W_p = \sum_{x_i \in \Omega} f_r(\|I(x_i) - I(x)\|) g_s(\|x_i - x\|)$$

'f(r)' is the range kernel and 'g(s) ' is the special.

Edge Enhancement

Our project uses Histogram Equalization (H.E) for edge enhancement. A histogram of an image is the graphical representations of intensity distribution of an image. It represents the pixel numbers in each colour component. H.E depends on the use of cumulative probability function(cdf), it is defined by [16]

$$cdf(x) = \sum_{k=-\infty}^{x} P(k)$$

To change the contrast of the edges or the image in general it shows the most frequent intensity values or stretches out the intensity range of the image, this would lead to the image's area of low contrast to have higher contrast.

Segmentation

After the dataset undergoes data pre-processing, we have used the "os" package in python to store the images. These images after pre-processing have to undergo segmentation. Firstly, we need segmentation in our project as it helps in acquiring the required white matter (WM) of the brain MRI. Brain tissue segmentation is a very challenging task due to heterogeneity in intensities, noise and different structures of the dataset. Tissues in the brain are CSF, WM, and GM they stand for cerebrospinal fluid (csf), white Matter (WM) and Grey Matter(CM).

K-Means Clustering

K-means set up for clustering is an unsupervised machine learning algorithm. It is very efficient in computing and gives satisfactoryiresultsiifitheiclustersiareicompactiandiwelliseparated The classification is performed by minimizing the distances between the data and the corresponding cluster centre. In K-means, all mage values are used to initialize the random centres and to calculate the Euclidian distance with the clustering centre. The algorithm is characterized by ability to segment image faster. Moreover, it reduces the computational complexity and improves the performances of K-means algorithm [18]. The results show that K-Means algorithm has good time complexity and accuracy. K-clusters are formed after partitioning the data based on the K-centroids..

K-means clustering steps

1. Select the number of clusters.
2. Then we take k as random points, and its centroids
3. Next we assign data to nearest centroid value, which in turn forms K-clusters.
4. After forming Clusters we compute new centroids of each new cluster.
5. The last step is to assign each datapoints closer to the centroids, if any reassignment occurs, go to step 4. Else, the model is said to be ready.

In our project, k-means clustering is used with the help of "Add packages" in python. In order to get the white matter (WM) and pass it into our model.

Modelling

This project uses CNN model in order to classify the image from one class to a different. CNNs are regularized variations of multilayer perceptron. Multilayer perceptron's commonly suggest completely related networks, this is, each neuron in one layer is attached to any or all neurons within the subsequent layer. The "full connectivity" of those networks causes them to be in danger of over fitting records. Regular methods of regularization, or preventing overfitting, consist of: penalizing parameters for the duration of education (consisting of weight decay) or trimming connectivity (skipped connections, dropout, and lots of others. In our problem statement we used Inception v3 because the architecture that classifies the test set into 4 classes. Inception v3 consists of 48 layers. It uses an input image size of 224X224 for best results. It uses 23 million trainable parameters and is an existing model of CNN architecture.

The Inception v3 model may be a resilient model because it can train hundreds and thousands of layers and still achieve great performance. The matter of exploding gradients was rectified by taking a shallow model and adding identity layers to it. This would provide a deep learning model that essentially must not produce higher training error than its counterpart because the layers added were only identity layers. The gradient exploding and training error problem was solved using this residual neural network where shortcut connections are used to perform identity mappings. This shortcut connection was needed because it helped in not having additional parameters being sent into the model and the computational time being very less.

IMAGE CLASSIFICATION
Our problem statement has images present in 4 classes. They are:
1. ND(non-demented)
2. VMD(Very mild-demented)
3. MD(mild demented)
4. MOdD(moderate demented)

The first class is the non – demented. It consists of 2560 images in training and 640 images in testing. The non-demented brain images provide a benchmark for other images to get classified. The second class is the Very mild – demented. This class consists of 2151 images in training and 448 images in testing. The characteristics of a very mild demented brain is a small decline in hippocampal region. The 3rd class is mild – demented. This class consists of 2548 images in the training set and 179 images in testing set. The characteristics of this class is little to small decline in hippocampal regions. The 4thcategory is the Moderate- demented. This class consists of 1792 jpeg files in the trainset and 12 jpeg files in the test set.

RESULT

The Inception v3 model is used to classify the demented images. The standard preciseness and assessments are used in detectingthe performance of categorization of models in this plot. About six evaluation metrics were used to evaluate the model performancefor the particular research, namely;

- Accuracy rate
- F1- Score
- Precision
- Recall
- AUC-ROC curve

A confusion matrix was generatedbased on the performance of this particular approach. [11] . It figured that, every confusion matrix contains the following four situations.

- **True positive (TP**): In this condition includes those patients who had the one in all 3 kinds of Alzheimer's or healthy brain. This also predicted that he is the one with the same situation.
- **True Negative (TN)**: This involves those who didn't have one in all 3 kinds of Alzheimer's .This predicts that patient is not having the same situation.
- **False positive (FP):**This condition includes those who didn't have one in all the 3 kinds of Alzheimer's .Nevertheless the prediction showed that patients had that scenario.
- **False Negative (FN**): It involves those patients who were one amongst the 3 styles of Alzheimer's or healthy brain. It conjointly expected without identical scenario.

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$$\text{Accuracy (ACC)} = \frac{TP + TN}{TP + TN + FP + FN}$$

$$\text{precision (PREC)} = \frac{TP}{TP + FP}$$

$$\text{recall (Sensitivity)} = \frac{TP}{TP + FN}$$

$$\text{f1score} = \frac{2TP}{2TP + FP + FN}$$

In our paper 9051 brain MRI images are categorized as about 2548 as mild demented, 2560 non-demented, moderate demented about 1792 and very mild demented 2151. The proposed diagnosed system stated in this paper correctly classifies and obtains 92.1% of accuracy. We also find out other factors like precision, F1-score and Recall. In our paper our proposed model is compared with different types of deep learning techniques, CNN and classical machine learning models. The Inception v3 's performance is compared to that of several approach withthe results which is presented in the composition publication. Our article uses Precision, accuracy, recall, F1-score and AUC of previous models which are then compared with proposed model. The method considered to acquire insights on the performance on both types of classification can be used. Our model antiquated on the MRI images which is our dataset. These are equated with the others, such as Hierarchical Extreme Learning Machine, multiclass deep learning method based on the Keras framework, Dementia Network (DEMNET). and results are provided in Table 3. The Inception v3 definitely performs better than all other models when we are comparing in terms of accuracy, recall and accuracy and we also see those results by classifying 4 different classes.

CONCLUSIONS

In this framework, the Inception v3 architecture is presented to identify Alzheimer's affected images. Convolutional layers and pooling layers with CNN are used in the Inception v3 architecture. The neural network method generates a pattern that can be classified as demented, mild demented, moderately demented, or non-demented. Following that, the k means segmentation process is performed to find the alzheimer's impacted pixels. The suggested method is validated using brain MRI images from an open-access dataset. The suggested method's performance metrics used are accuracy, precision, F1-score, and recall. The suggested alzheimer's detection obtains an accuracy rate of 92.26 %, with a loss of 0.2117, precision of 94.6%, F1-Score of 92.1 percent, and recall of 95.5 percent.

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Table 1 : Data Before and after augmentation

	Test Data	Train Data	
		Before Augmentation	After Augmentation
Non-Demented	640	2560	2560
Mild Demented	179	52	2548
Moderate Demented	12	1792	1792
Very Mild Demented	448	717	2151

Table 2 : Model comparison

Ref	Dataset	Modality	ACC	PR	REC	F1-Score
MLP[21]	ADNI	MRI	89	85	87	89
CBLSTM[21]	ADNI	MRI	82	79	82	82
CBLSTM +SMOTE [21]	ADNI	MRI	82	78	88	82
DEMNET[22]	Kaggle	MRI	85	80	88	83
H-ELM [22]	Kaggle	MRI	82	79	81	82
Proposed Method	Kaggle	MRI	92.26	94.6	95.5	92.5
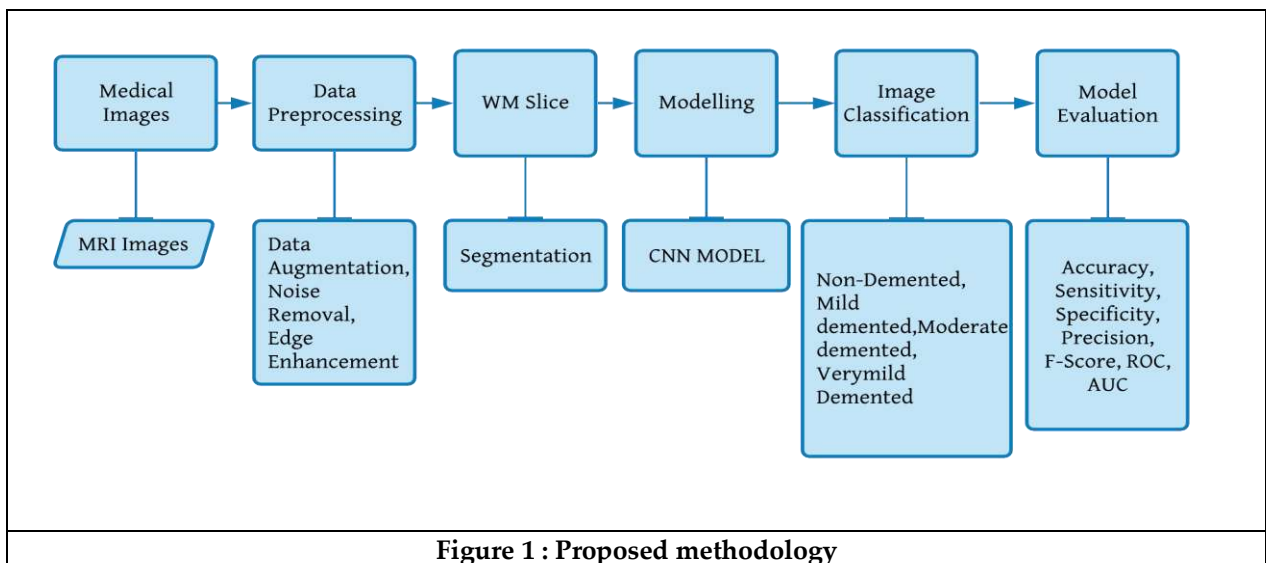
Figure 1 : Proposed methodology

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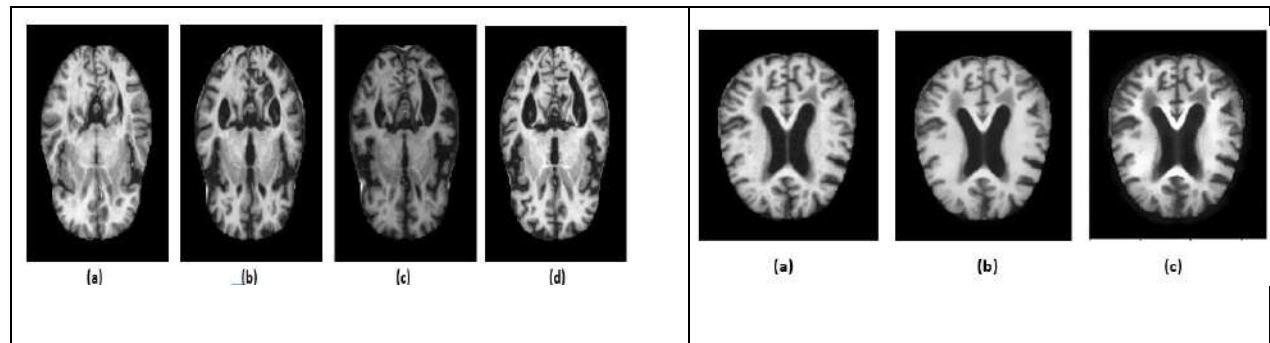
Figure 2 : (a) ND (b) VMD (c) MD (d) MODD

Figure 3: (B) First image (B) After pre-processing (C) Edge enhancement done
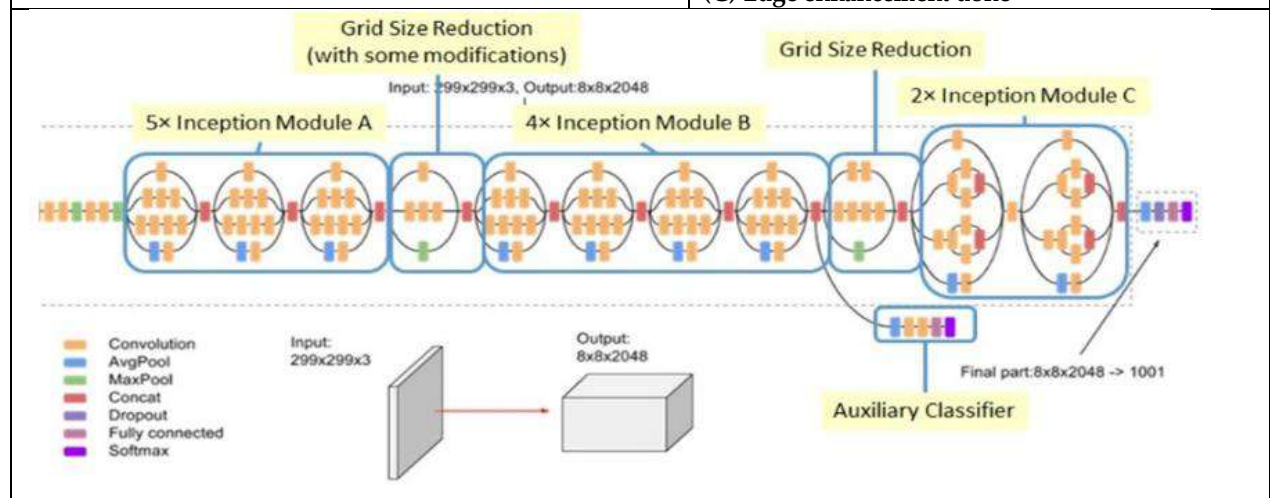
Figure 4: Architecture of Inception v3
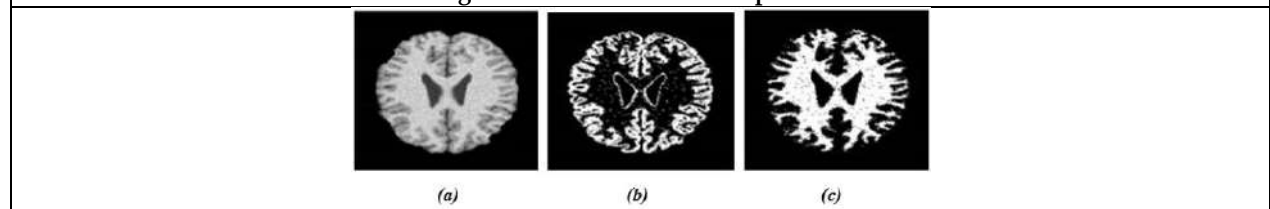
Figure 5 : (a). Original image (b). Gray matter (c). White
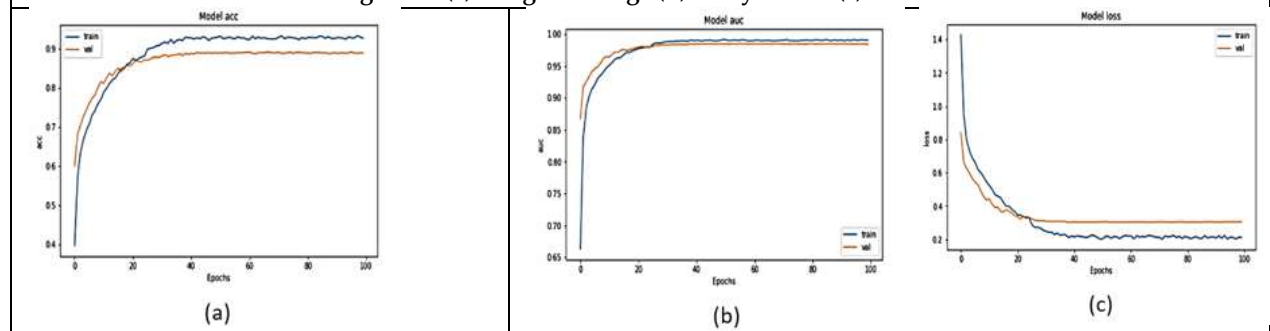
Figure 6 : Evaluation plots (a) Accuracy   (b) AUC   (c) loss

RESEARCH ARTICLE

Structural, Morphological Characterization and Antibacterial Activity of Mahavat Vidhwansan Rasa

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ABSTRACT

Herbal medicines based on Ayurveda formulations are very important for the treatment of many types of diseases. In this study structural and morphological characters of synthesized Mahavat Vidhwansan Rasa have been investigated. Most of the herbal-based formulations are currently used as antimicrobial agents. Mineral-based Rasa medicine is used extensively in Ayurvedic literature for the management of various diseases like Vata dosha, anemia, piles, epilepsy, etc., as iron, tin, copper are very hard metals, these metals have been converted into Rasa (herbo metallic preparation) for traditional medicinal use. The Ayurvedic drug has been characterized using grazing angle X-ray diffraction (XRD), Scanning electron microscopy (SEM), and optical absorption spectroscopy. In this study the most prominent peak indicates the presence of SnO_2 or Vanga bhasma using X-ray diffraction; similarly Scanning electron microscopy represents the nano- cluster formation of metal complexes. The highly effective Ayurvedic herbo-mineral drug Mahavat Vidhwansan Rasa has been investigated for antibacterial activity against some pathogenic bacterial species. Agar well diffusion method was used for the analysis of antimicrobial activity. The results of the present study indicate that Mahavat Vidhwansan Rasa is shown to exhibit promising antimicrobial activity against the test bacterial species.

Keywords: Herbo metallic preparation, Mahavat Vidhwansan Rasa, XRD, SEM,UV-Vis, Antimicrobial property, Agar well diffusion.

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INTRODUCTION

The explanation of Mahavat Vidhwansan Rasa is present in chapter 37 Kharaliya Rasayanadhyaya of Rasashastra Siddha prayoga sangraha [Sharma *et al.* 2019]. The main ingredients of Mahawat vidhwansan rasa are Parada shuddha, Gandhaka shuddha, Nagabhasma (shataputi), Vanga bhasma, Lauha bhasma, Tamra (bhasma), Abhraka (bhasma), Pippali, Saubhagya (Shuddha Tankan), Maricha, Shunthi each of one part alomg with this four-part of Vatsanabha (shuddha). Then bhavana of Trikatu kvatha, Triphala kvatha, Chitraka mula kvatha, Bhringaraja svarasa, Kushtha kvatha, Nirgundi Patra svarasa, Arka dugdha, Amalaki svarasa, Ardraka svarasa, Nimbu svarasa each of 3 times. It is recommended in the dose of 125 mg or 1 ratti in once or twice a day dose along with anupana of Ardraka svarasa (ginger juice), Bhringaraja svarasa (Eclipta Alba juice), Eranda Taila (castor oil), Ghrita (Ghee) or Madhu (Honey) depending upon the age, digestion, strength and the condition to be treated. As it contains minerals like Mercury, and Sulphur, Lead, Tin, Iron, Copper, Mica, so the proper shodhana process should be followed during the manufacturing of Mahavat Vidhwansan Rasa. If not followed by shodhana processes it may cause toxic symptoms. One of important ingredients of Mahavat Vidhwansan Rasa is Vatsanabha (Aconitum ferox), it may causes side effects like Blackout, Bursting type of pain over the back of the head, Cold hand, and feet, Heaviness of tongue, Irregular rhythm low volume, Low blood pressure, and pulse rate, Nausea, Numbness and tingling sensation of palm and sole, Numbness around the mouth, Retention of urine, Severe sweating, Vertigo, etc. if taken in higher doses. With this object, the Mahavat Vidhwansan Rasa was prepared and analyzed as nanoclusters for quality testing or quality control, on the parameters described in ayurvedic literatures as well as advanced science and technology.

MATERIALS AND METHODS

Material Synthesis and Experimental Methods
The raw material of bhasams (mineral-based) was taken as raw material for the Rasa synthesis process. The preparation involves mainly two steps- normal purification (Shodhana) [1], special purification (titration through herbal extracts), in which a modern synthesis technique is used to produce the final product, Mahavat Vidhwansan Rasa [Sharma VK, *et al.* 2019].

Ingredient Specification

X-ray Diffraction
X-ray diffraction images were recorded using Bruker D8 diffractometer in grazing angle mode at an angle of incidence of 0.5^o. Wide-angle X-ray spectra were recorded in the 2θ range of 20° to 60° with a step size of 0.02^o.

Spectroscopic Analysis
Optical absorption experiments were carried out to study quantum confinement in the Ayurvedic herbo-mineral drug nanoparticles using dual-beam Toshwin Shimadzu (UV-1800) supplied by Shimadzu (Japan) [Drbohlavova, *et al.* 2009] Photoluminescence measurements were carried out at room temperature using a Perkin Elmer LS-55 spectrometer within the spectral range of 190 to 650 nm with $\lambda_{accuracy}=\pm0.5$ nm.

Morphological Examination by Scanning Electron Microscope
The morphological examination of the Ayurvedic herbo mineral drug was studied with the help of a scanning electron microscope JSM-5600.

Antimicrobial Activity of Mahavat Vidhwansan Rasa

Test Microorganisms for Antimicrobial Activity

Escherichia coli, Streptococcus pyogenes, Staphylococcus aureus, Pseudomonas aeruginosa and *Salmonella typhi*. [Bhalodia and shukla *et al.* 2011]

Preparation of DMSO solutions of Mahavat Vidhwansan Rasa

The Mahavat Vidhwansan Rasa was diluted to obtain the solution at the desired concentration. 20% DMSO   was used as diluents to get desired concentration of drugs to test upon standard bacterial strains. Three solutions of 50 mg/ml, 75 mg/ml, and 125 mg/ml were prepared by mixing 20%   DMSO   Mahavat Vidhwansan Rasa.

Antimicrobial Susceptibility Testing

Antimicrobial activity was detected by agar well diffusion test with some modifications [Gulluce *et al.* 2007]. In this method, Mueller -Hinton agar media was prepared and the test organisms were aseptically inoculated onto the surface of the media using a sterile swab. Agar wells were prepared with the help of a sterile Pasteur pipette borer. Dilutions of Mahavat Vidhwansan Rasa, DMSO-20% Solution and Streptomycin were added to the wells of the inoculated plates. The plates were incubated in an upright position at 37±1°C for 24 hrs [Rawat and Swarup, 2015]. The extract diffuses into the agar media and inhibits the growth of microbial strain-producing zone of Inhibition. The zone of inhibition (including the diameter of the antibiotic) was measured and expressed in millimeters (mm) [Trigui *et al.* 2013].

RESULTS AND DISCUSSION

X-Ray Diffraction Studies

The as-prepared Ayurvedic herbo mineral drug Mahavat Vidhwansan Rasa nanoparticles were also characterized by X-ray diffraction. The observed diffraction patterns of the drug sample have been shown in Figure 2. The experimentally determined (h,k,l) planes and assigned phase have been compared with the standard JCPDS data in Table 2. The Naag bhasma (Pb_2O_3) exhibited four peaks at 2θ=23.44°, 31.71°, 41.21° and 54.37° as shown in Figure 2, which were assigned to the (001), (020), (211) & (031) planes of the monoclinic phase of Pb_2O_3. Nagarajan et al have also observed the presence of the monoclinic phase in Naag bhasma samples [Nagarajan *et al.* 2012]. The X-Ray diffraction spectra recorded for Mahavat Vidhwansan Rasa were marked by three broad peaks centered at 26.67°, 33.52°& 52.16°. The first most prominent peak has a Gaussianline shape, which indicated the presence of a Rutile phase of Vang bhasma (SnO_2). The second and third observed peaks, however, were asymmetric. The asymmetry in the line shape of this peak may arise due to the merging of two closely spaced reflections due to a reduction in crystalline size. The most prominent peak in the Mahavat Vidhwansan Rasa X-Ray diffraction spectra was due to the SnO_2 (110) plane parallel to the substrate surface while the other peaks were relatively weak in intensity. This indicated that the synthesized SnO_2 samples grew with a preferred (110) orientation. The average value of the lattice constant was found to be a = b = 0.4722 nm and c = 0.3124 nm, which match well with the standard XRD JCPDS data file of SnO_2 [Kelp *et al.* 2016]. Rudramma and Narang, [Hiremath and Narang. 2010] also studied the XRD Analysis of Vanga Bhasma and identified XRD peaks to be as Tin dioxide (SnO_2). Swamy and Ravikumar, [Swamy and Ravikumar. 2010] also studied X-ray diffraction of Ayurvedic medicines and analyzed 22 elements from the ayurvedic medicine.

The mean particle size of Mahavat Vidhwansan Rasa sample was estimated from the X-ray diffraction pattern by using the three most prominent peaks according to the scherrer equation.

$$d = \frac{0.9\lambda}{B\cos\theta}$$

Where B = $\sqrt{B_m{}^2 - B_S{}^2}$ is relative broadening (FWHM).

λ is the wavelength of CuKα (1.5402 nm), 2θ diffraction angle, B_m is measured broadening and B_s is the standard broadening determined for standard silicon sample. Our XRD data also overrule the possibility of the formation of a ternary alloy phase of the type SnO_2 since it should also manifest as peak narrowing. Since the (110) peak half-width remained practically constant, we can conclude that the XRD data affirm the formation of the Sn/Pb core-shell structure.

Optical Absorption Spectroscopy
The optical absorption spectra of Mahavat Vidhwansan Rasa sample with different particle sizes have been shown in the figure. Note that for each sample, the sharp rise in the absorption onset corresponds to the fundamental absorption edges which exhibit systematic blueshift with the reduction in particle size. The corresponding spectral position of fundamental absorption edges has also been marked. The optical absorption onset could be assigned to transition from the lowest occupied ($1S_h$) to the highest unoccupied ($1S_e$) levels. Obviously, the SnO_2 and iron oxide quantum dots exhibit large blue shifts in the corresponding absorption edges due to the quantum confinement effect [Kaushik *et al.* 2006].

Scanning Electron Microscopy
Figure 3 represents the scanning electron micrograph of Mahavat Vidhwansan Rasa sample at 30 min of deposition time. It is clear from the figure that, the core-shell sample after 30 min of deposition are seen to form Pallet type clusters. The Scanning Electron Microscopy images from the self-assembled thin film after 60 min of deposition time gave rise to another interesting feature with microbead type cluster observed from the SEM image [Morillas *et al.* 2015]. The average cluster size was measured from the SEM images and the particle size histogram is shown in the inset Figure 4. The average cluster size was measured as 250±5 nm which is in good agreement with XRD. The present study is also in agreement with the findings of Virupaksha and Kumar [Virupaksha and Kumar. 2012] with their study based on the structural analysis of Tarakeshwara Rasa by Scanning Electron Microscopy.

Anti Microbiological Studies
The present study was conducted to analyze the antibacterial activity of prepared Mahavat Vidhwansan Rasa. The results in this study were analyzed in terms of the diameter of the zone of inhibition and recorded as antimicrobial activity [Khan *et al.* 2019]

Different concentrations Mahavat Vidhwansan Rasa was prepared for analysis of antibacterial activity. The results were compared to the commercially available antibacterial drug Streptomycin. The study indicates that Mahavat Vidhwansan Rasa exhibited good antimicrobial activity against *Escherichia coli, Streptococcus pyogenes, Staphylococcus aureus, Pseudomonas aeruginosa* and *Salmonella typhi.* At 50 mg/ml concentration, Mahavat Vidhwansan Rasa inhibited the growth of *Escherichia coli* and *Staphylococcus aureus* but was not effective against *Streptococcus pyogenes, Pseudomonas aeruginosa,* and *Salmonella typhi.* In this study, the best combination to inhibit the growth of bacteria was found to be a 125 mg/ml concentration of Mahavat Vidhwansan Rasa. The results of the present study indicate that prepared Mahavat Vidhwansan Rasa was equally effective as the standard antimicrobial drug Streptomycin. [Wang *et al.* 2017]

Maximum zone of inhibition was found in the plates of *E.coli* (30 mm) at the concentration of 125 mg/ml. *E.coli* was found to be the most sensitive bacterial species for Mahavat Vidhwansan Rasa at all concentrations and *Pseudomonas aeruginosa* was analyzed at least sensitive bacterial species in terms of zone of inhibition and antibacterial activity. Data are means of three replicates [Khan et al., 2019]. The results of the present study are in agreement with the findings of Tambekar and Dahikar [Khan and Bhadauria. 2017] with their study on antibacterial activity of Ayurvedic medicines against enteric pathogens.

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CONCLUSION

The traditional Indian method of synthesizing Rasa (Oxidenanoparticles) was successful, which was observed from spectroscopic studies. By undergoing normal (Shodhan) and special purifications (titration with herbal extracts or bhavna dravya) the particles of Rasa aushadhi were tuned into nanocrystals in some concentration. The most prominent peak in the X-Ray diffraction spectra of Mahavat Vidhwansan Rasa was due to the SnO_2 (110) plane parallel to the substrate surface while the other peaks for iron, copper & lead oxides were relatively weak in intensity. Excellent antimicrobial activity was observed in Mahavat Vidhwansan Rasa where the maximum zone of inhibition up to was observed which was better than all standard drugs. The application of heavy metal-based herb mineral nanoparticles in the field of antimicrobial activity is still in its early days.

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Disclosure of Any Conflict Of Interest
The author declares that there is no competing interests or personal relationships that could have appeared to influence the work reported in this paper.

Abbreviation
XRD: X-ray diffraction, **SEM:** Scanning electron microscope, **UV-Vis:** Ultraviolet–visible spectroscopy, **TEM:** Transmission Electron Microscope

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Table.1. Ingredients names of Mahavat Vidhwansan Rasa

S.No	Ayurvedic Name	Common Name
1	Shuddha Parada	Herbal purified Mercury
2	Shuddha Gandhaka	Herbal purified Sulphur
3	Naga bhasma	Lead Calx
4	Vanga bhasna	Tin Calx
5	Loha bhasma	Bhasma prepared from Ironn
6	Tamra bhasma	Bhasma prepared from Copper
7	Abhraka bhasma	Purified and processed Mica
8	Pippali	Long pepper fruit-pipper longum
9	Tankana bhasma	Borax
10	Maricha	Black pepper-Piper longum
11	Shunti	Ginger Rhizome-Zingiber officinalis
12	Shuddha Vatsanabha	Purified Aconitum ferox
13	Trikatu	Pepper, long pepper, and ginger
14	Triphala kwath	
15	Chitrakamoola	
16	Bhringaraja	Eclipta alba
17	Kushta	Saussurea lappa
18	Nirgundi	Vitex negundo
19	Arkaksheera	Latex of Calotropis procera
20	Amalaki	Indian gooseberry fruit – Emblica officinalis Gaertn.
21	Ardraka	Ginger Rhizome – Zingiber officinalis
22	Nimbu Swarasa	-Nimbu Swarasa

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Table2. Summary of the XRD peak positions (2θ), corresponding (hkl) value, different compounds, and crystalline phase for Mahavat Vidhwansan Rasa

Compound Name	Sample	2θ (degree)	(hkl)	Phase Assin
Mahavat Vidhwansan Rasa	Pb_2O_3	23.44	(001)	monoclinic Pb2O3 [JCPDS 76-1791]
		31.71	(020)	
		41.21	(211)	
		54.37	(031)	
	SnO_2	26.67	(110)	Rutile JCPDS-041-1445
		33.52	(101)	
		52.16	(211)	
	CuO	39.07	(111)	Monoclinic JCPDS No 48-1548
		49.85	(202)	
	Fe	44.19	(110)	Cubic, JCPDS No. 87-0721
	Fe_3O_4	35.92	(311)	Cubic, JCPDS No. 65-3107

Table 3. Antibacterial activity of Mahavat Vidhwansan Rasa sample

The concentration of Mahavat Vidhwansan Rasa /Standard	Zone of Inhibition				
	Escherichia coli	*Streptococcus pyogenes*	*Staphylococcus aureus*	*Pseudomonas aeruginosa*	*Salmonella typhi*
50 mg/ml	14	ND	11	ND	ND
75 mg/ml	20	12	14	ND	ND
100 mg/ml	25	18	21	13	14
125 mg/ml	30	23	26	19	21
Streptomycin (10mcg)	18	15	16	11	16
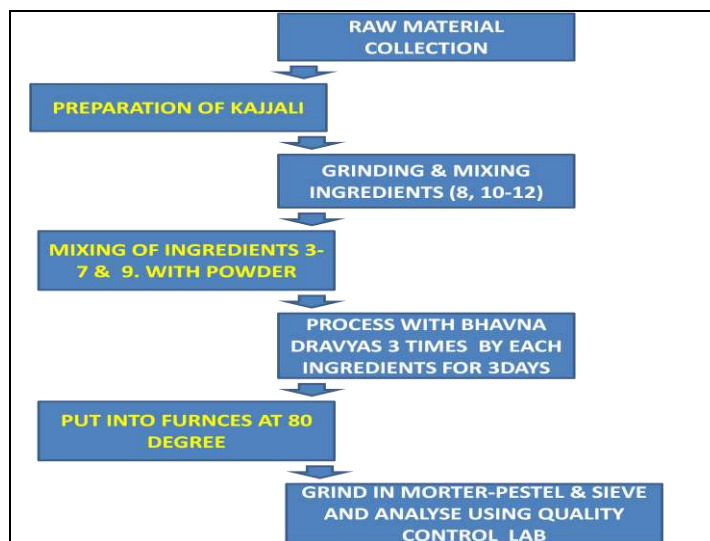
Figure 1. Manufacturing process diagram Mahavat Vidhwansan Rasa
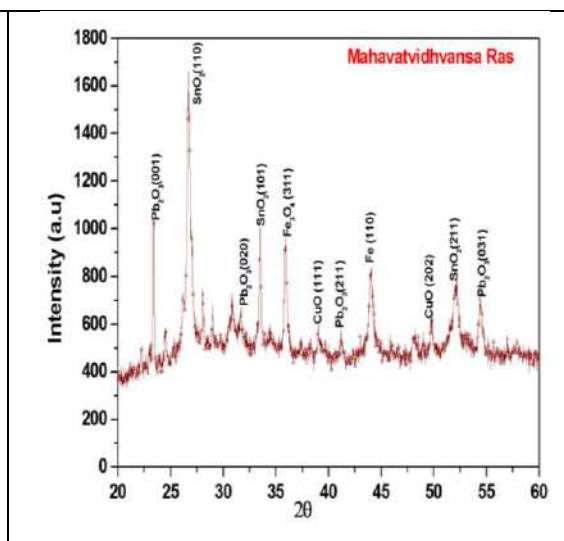
Figure 2. Grazing angle X-Ray diffraction spectra of Mahavat Vidhwansan Rasa
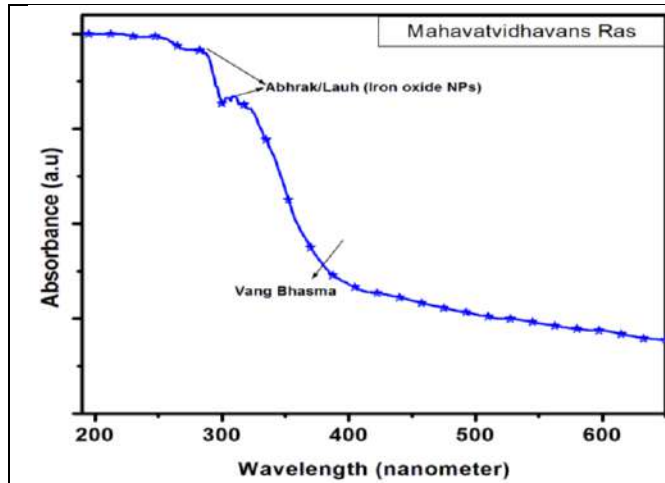
Figure 3. Optical absorption spectra of Mahavat Vidhwansan Rasa
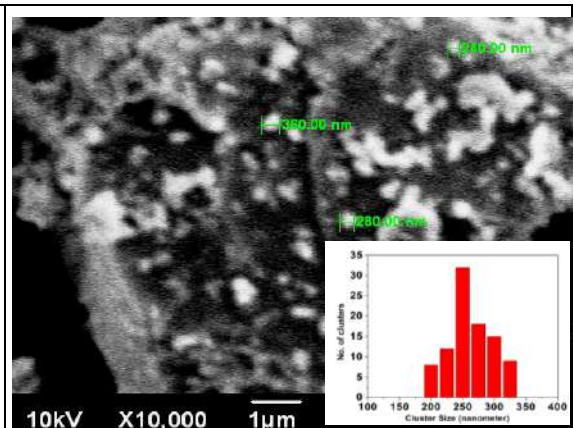
Figure 4. Scanning electron micrograph of Mahavat Vidhwansan Rasa. The lower inset figure represents cluster size histogram or particle size distribution
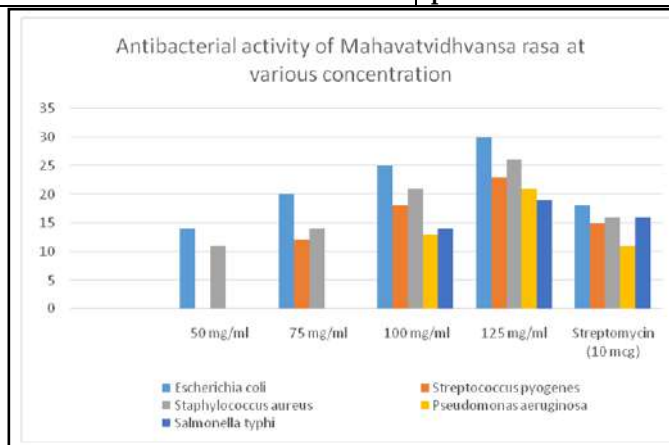
Figure 5. Antibacterial activity of Mahavat Vidhwansan Rasa at various concentrations

RESEARCH ARTICLE

Fatty acid Composition of Indian Pearl Spot (*Etroplus suratensis*): A Comparative Study between Brackish and Freshwater of Vembanad Wetland in Alappuzha, Kerala

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ABSTRACT

Indian pearl spot (*Etroplus suratensis*) is one of the most consumed species among freshwater as well as brackish water reared fishes in southern part of India. However, studies show the percentage comparison of fatty acids composition in both fresh and brackish water fish. The chemical composition of the samples was determined using AOAC methods. We found that the two different ecosystem (fresh and brackish water) fish species studied had high protein-medium oil (brackish water), whereas fresh water had high protein-low oil. They are high in fatty acids and total proteins. Fish species can be used liberally in diets to avoid excessive consumption of saturated fat, which, if not supplemented with polyunsaturated fats (PUFA), will be utilised to create cell membranes. However, the resultant cell membranes are highly elastic, and good to the heart. As a result, both freshwater and brackish water fish species include important fatty acids, cholesterol, and proteins that promote good health, illness prevention, and healing in humans. On the contrary, brackish fishes were discovered to be more nutrient rich than freshwater fishes.

Keywords; *Etroplus suratensis*, brackish and water fishes, polyunsaturated fats, cholesterol, proteins contents.

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INTRODUCTION

Fishes are an excellent source of nutrition, as well as being delicious and easy to digest. More than a one third of the population in developing countries get their animal protein from fishes. Fishes are particularly rich in the amino acid (lysine), which is found in substantial levels in comparison with cereals (Ye et al., 2017). Polyunsaturated fatty acids (PUFA) found in fish oils, such as linoleic and arachidonic acids, are known as "essential" fats. In addition, fish and fish oils have been shown to have an important role in enhancing brain development and lowering the risk of cardiovascular disease. In addition to working directly on the growth process and human development, the consumption of fish oils containing PUFA lowers the biochemical variables linked to cardiovascular disease, diabetes, rheumatoid arthritis, and cancer (Simopoulos, 1991).Eicosapentaenoic acid (EPA) is one of these fatty acids that has gotten a lot of interest recently when Danish researchers discovered that the diets of Greenland Eskimos contain substantial amounts (Elvevoll and James, 2002).Essential fatty acids linoleic acid and linolenic acid are metabolised sequentially by denaturation and elongation enzyme systems, resulting in the formation of Light chain-PUFA. Likewise, Freshwater fish may also denaturize their fatty acid chains to generate EPA and arachidonic acid, which are then elongated to produce DHA and docosapentaenoic acid (Parrish, 1999).Fishes muscle is the primary portion consumed by humans, thus its fatty acid and proteins makeup should be taken into account when recommending it as a health food. Many cardiologists advise their patients to eat enough of fish to avoid consuming too much saturated fat and to get enough protein in their diet without consuming too much fat, which can lead to weight gain. As a result, the health benefits of fish lipids and proteins have been widely accepted (Ackman, 1990).

The Indian pearl spot (*Etroplus suratensis*, Cichlidae), native to Asia, cultivated in the Indian state of Kerala and also in Sri Lanka, which has a high market demand and a large body size, making it a good prospect for aquaculture. For aquaculture promotion, it was introduced to several Indian states along the eastern and west coasts. This fish, which was designated as Kerala's official state fish in 2010, is a major food source and a top choice for brackish and freshwater aquaculture in India.Fishers interested in key fish species like Indian pearl spot should be aware of their fatty acid content, not only because fish are valuable products, but also because fish oil may have new industrial uses. The fatty acid content of this group of fish is significant in view of recent nutritional and medical attention to the significance of fatty acids in human physiology.Many scientist has written about pearl spot commercial cultivation in India's several agro-climatic areas (Padmakumar et al., 2012; Sukumaran et al., 2017; Peter et al., 2019). Long-chain n-3 polyunsaturated fatty acids (LC n-3 PUFA), particularly eicosapentaenoic acid (EPA) and docosahexaenoic acid (DHA), are abundant in fish lipids (DHA). Humans cannot synthesise long chain, n-3 PUFA and must get them through diet(Alasalvar et al., 2002). According to several researches, PUFA are known to impact prostaglandin production and hence improve wound healing (Bowman and Rand, 1980; Gibson, 1983). Polyunsaturated fatty acids (PUFA) have been proven to have beneficial effects on cardiovascular disease and cancer (Connor, 1997). Thus, it is vital for human health to increase consumption of polyunsaturated fatty acid-rich fish and fish products (Sargent, 1997). The content of PUFA varies between fish species, even between freshwater and marine fish (Rahnan et al., 1995).

The lipids, protein, and fats present in various fish species have been widely studied in temperate regions across the world. The most important species, pearl spot, was chosen for this study because it has high consumer acceptance, is commercially viable, and has a low fat content. They are also the most farmed fish in southern India, and as a source of relatively affordable animal fat, they have become an increasingly significant component of the nation's nutrition. The literature, however, contains no information on their nutritional composition. Despite the high demand, commercial value, and widespread distribution of these fish, precise nutritional data on their nutritional composition is needed in South-west India, where they are frequently consumed. As a matter of fact, the goal of this study was to look into the proximate fatty acid content of the *Etroplus suratensis* (Pearl spot) fish species in freshwater and brackish water.

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MATERIALS AND METHODS

The present work was conducted at the highly brackish and Fresh water of Vembanad Lake in the district of Ernakulum, Kerala during 2018-2019. Live specimens of *Etroplus suratensis* were purchased while samples were stored in an ice chest at 4^{0}C and conveyed to the laboratory. The specimens were packaged in separate labelled polythene bags and eventually stored in a cold store (-22°C) at the Department of Zoology, Bangalore University Bengaluru. Fish samples were thawed in the open air in the laboratory and individual data for length, weight and sex taken and recorded. The standard length was measured using a measuring board. The weight was measured with an electronic weighing balance. Each fish was dissected, gutted and the gonad removed to determine the sex by visual examination. The fish sample was then cleaned, filleted and placed in a Waring Blender and homogenized for 15 min. Samples for the different chemical analyses were then taken from the homogenous material. Triplicate determinations were carried out on each sample. The fats were converted to free fatty acids by saponification. The fatty acids were converted to their methyl esters and into heptane. Internal standards were used for estimation of actual fatty acids present in the fat. Identification/quantification of fatty acids was achieved by gas chromatography, the former being resolved by elution times. The analysis was carried out by a commercial analysis laboratory.

Statistical analysis
The descriptive statistics (mean ± standard error) were conducted while statistical significance of differences (P≤ 0.05) was determined by analysis of variance (ANOVA) with SPSS version 20.0

RESULTS

The average length of the adults *Etroplus suratensis* in brackish water were109.95 ± 5.62,and 180.05 ± 13.28 were from fresh water, and the average weight of *Etroplus suratensis* (180.33 ± 12.72 and 108 ± 3.61) in both fresh and brackish water respectively (Table 3).

Protein content
The mean percentage of protein content of *Etroplus suratensis*in fresh water fishes were19.5±0.4while that of brackish waterwas18.9±0.1. No significant difference was noted (p>0.05) between the ecosystem. The highest protein content was recorded on fresh water (Table 1).

Lipid, Carbohydrate and Cholesterol content
The mean percentage of lipid value of Fresh water from 0.5±0.2 and that of brackish water from 1.3±0.4. No significant difference was noted (p>0.05) between the ecosystem. The highest lipid content was obtained on the samples from brackish water. However, Carbohydrate content in both fresh and brackish water was found to be negligible(Table 1). The mean percentage of Cholesterol content of *Etroplus suratensis* in fresh water fishes were 53.0±0.2while that of brackish water was 65.7±0.2. No significant difference was noted (p>0.05) between the ecosystem. The highest protein content was recorded on brackish water(Table 1).

Fatty acid content
Table 2 summarises the fatty acid content of the *Etroplus suratensis* species as a percentage of eluted methyl esters in both brackish and fresh water. The order of the fatty acids is determined by their chromatographic retention durations. Among these, myristic acid was found in the greatest concentrations (7.2±0.2 and4.3±0.4%), palmitic acid (27.9±0.2 and40.0±0.4%), stearic acid (35.0±0.2 and22.9±0.4%), oleic acid (26.3±0.1 and21.2±0.4%), linoleic acid (77.3±0.2 and1.8±0.4%), Linolenic acid (3.3±0.2 and 0%), Arachidonic acid (44.4±0.2and0%), Behenic acid (3.7±0.3 and1.6±0.5%), Erucic acid % (6.9±0.2 and 0%), Lignoceric acid (2.8±0.2 and 0%), Recenoleic acid (3.7±0.2and 0%), Capric acid (44.0±0.2 and 0%), and Lauric acid (2.2±0.2 and1.5±0.4). When comparing the two ecosystems, brackish water fishes have more fatty acids than fresh water fishes.

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DISCUSSION

Like many animals, fish have the potential to accumulate lipids in their bodies. There are several classifications that can be done by dividing fish into several groups, for example consistent with their lipid content. *Etroplus suratensis* belongs to the high protein-medium fat in fresh water group, whereas brackish water contains high protein-low fat (Kleimenov, 1971). According to Osman et al., (2001), low-fat fish have greater water content, whereas total fat content in fresh water was lower by weight, while brackish water was higher.Also, species, geographical area, age, and diet all have an impact on fat content (Piggott and Tucker, 1990). In general, the fatty acid content of the fish studied agrees with earlier studies on the fatty acid composition of comparable fish species (Jabeen and Chaudhry, 2011). Palmitic acid was found to have the highest amount of saturated fatty acids in the two distinct ecosystems studied. Arachidonic acid, a precursor for prostaglandin and thromboxane production, was found in brackish water fish species (Pompeia et al., 2002). Although the amount of arachidonic acid was in a significant range, it can interfere with the blood clotting process and bind to endothelial cells during wound healing (Rahnan et al., 1995). This, however, contradicted the findings in three Malaysian Channa spp fish (Zuraini et al., 2006). Cholesterol levels in the current study show that both the fresh and brackish water fish species may have a therapeutic impact on muscular soreness and inflammation, and this was in agreement with the Mohsin, (1983). Cholesterol has been shown to inhibit wound healing in both roadents and humans (diabetes individuals with foot ulcers) (Chen et al., 2021; Kim et al., 2010). As a result, fish has been proposed as an important component of a healthy diet in humans (Rahnan et al., 1995).

The unsaturated fatty acids (UFA)has been proposed as a helpful measure for evaluating the relative nutritional benefits of different fish oils. As previously observed, the UFA ranged more in brackish water fishes than in fresh water fishes, showing significant species diversity (Hearn et al., 1987). The PUFA/total lipid ratio of freshwater fish was considerably lower than that of marine fish. It was proposed that it would make up a healthy human diet(Kleimenov, 1971).The UFA levels in the freshwater fish were within the acceptable range.Although it is widely acknowledged that UFA content varies between fish locations, little attention has been devoted to the UFA composition of various ecosystems when selecting fish for diets. All fish were thought to have comparable nutritional value, and selection was based largely on availability, freshness, flavour, and other similar criteria (Hearn et al., 1987). According to the findings of this study, cholesterol and fatty acids, which are exclusively present in brackish water fish, have highly favourable qualities for the prevention of human coronary artery disease, and this was in accordance with (Goodnightet al., 1982). Antithrombotic dietary supplements based on fish oils have been proposed (Goodnightet al., 1982). As a result, while suggesting fish for health promotion, both the fat content and the UFA distribution must be taken into account. The current study found that brackish water fish species contained large amounts of cholesterol and UFA, making them an average source of polyunsaturated fatty acids (PUFA).

CONCLUSION

We found that the two different ecosystem (freshwater and brackish water) fish species studied had high protein-medium oil (brackish water), whereas fresh water had high protein-low oil. They are high in fatty acids and total proteins. Fish species can be used liberally in diets to avoid excessive consumption of saturated fat, which, if not supplemented with polyunsaturated fats, will be utilised to create cell membranes. However, the resultant cell membranes are highly elastic, and good to the heart. As a result, both freshwater and brackish water fish species include important fatty acids, cholesterol, and proteins that promote good health, illness prevention, and healing in humans.On the contrary, brackish fishes were discovered to be more important nutrient suppliers than freshwater fishes.

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Table 1; A study on biochemical composition (proteins, fats, carbohydrates and cholesterol) of Indian pearl spot (*Etroplus suratensis*) fishes in fresh and brackish water

Parameters		Mean±SEM
Protein (g/100g)	Brackish Water	18.9±0.1
	Fresh Water	19.5±0.4
Fat (g/100g)	Brackish Water	0.5±0.2
	Fresh Water	1.3±0.4
Carbohydrate (g/100g)	Brackish Water	0.0±0.0
	Fresh Water	0.0±0.0
Cholesterol (mg/100g)	Brackish Water	65.7±0.2
	Fresh Water	53.0±0.2

Values are Mean ±SEM, of three separate determinations

Table 2; Fatty acid composition of Indian pearl spot (*Etroplus suratensis*) fishes in fresh and brackish water

Parameters		Mean±SEM
Caproic acid %	Brackish Water	0.0±0.0
	Fresh Water	0.0±0.0
Caprillic acid %	Brackish Water	0.0±0.0
	Fresh Water	0.0±0.0
Capric acid %	Brackish Water	4.0±0.2
	Fresh Water	0.0±0.0
Lauric acid %	Brackish Water	2.2±0.2
	Fresh Water	1.5±0.4
Myristic acid %	Brackish Water	7.2±0.2
	Fresh Water	4.3±0.4
Palmitic acid %	Brackish Water	27.9±0.2
	Fresh Water	40.0±0.4
Stearic acid %	Brackish Water	35.0±0.2
	Fresh Water	22.9±0.4
Oleic acid %	Brackish Water	26.3±0.1
	Fresh Water	21.2±0.4
Linoleic acid %	Brackish Water	7.3±0.2
	Fresh Water	1.8±0.4
Linolenic acid %	Brackish Water	3.3±0.2
	Fresh Water	0.0±0.0
Arachidonic acid %	Brackish Water	4.4±0.2
	Fresh Water	0.0±0.0
Behenic acid %	Brackish Water	3.7±0.3
	Fresh Water	1.6±0.5
Erucic acid %	Brackish Water	6.9±0.2
	Fresh Water	0.0±0.0

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Lignoceric acid %	Brackish Water	2.8±0.2
	Fresh Water	0.0±0.0
Recenoleic acid %	Brackish Water	3.7±0.2
	Fresh Water	0.0±0.0

Values are Mean ±SEM, of three separate determinations

Table 3; Morphometric analysis ofIndian pearl spot (Etroplus suratensis) fishes in fresh and brackish water.

Parameters		Mean±SEM
Weight(g)	Brackish Water	108.00±3.61
	Fresh Water	180.33±12.72
Length (cm)	Brackish Water	109.95±5.62
	Fresh Water	180.05±13.28
Width (cm)	Brackish Water	47.00±2.71
	Fresh Water	94.76±6.99

Values are Mean ±SEM, of three separate determinations

RESEARCH ARTICLE

Protein Functional Annotation and Network Analysis in Relation to Oral Pathogen and Oral Cancer using Computational Tools

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ABSTRACT

Oral cancer is becoming more common, and it threatens to be a serious worldwide medical issue. Approximately 100 trillion microorganisms' cells live in harmony with their host in the human body. Bacteria have long been thought to have a role in immunological regulation, disease onset, and maintenance of optimal health at certain body locations. This study aims to elucidate the networks between proteins and biologically active compounds, as well as their functional annotations, cell signaling pathways. According to the results, online STRING software was used to create a molecular genetic interaction network azurin on oral bacterial proteins, and Cytoscape software was used to calculate 11 nodes, 16 edges, and an average node order of 2.91 and average value. The local clustering factor is 0.876, the expected number of edges is 10, and the enrichment ppi has a p-value of 0.0553. Nodes, lines and colors prove the rationality of interactive networksFor interleukin 8 in oral squamous cell carcinoma, a molecular genetic interaction network is obtained and Cytoscape software is used to visualize the number of nodes 11, the number of edges 55, the average node order 10, and the average. Did. The local clustering factor is 1, the expected number of edges is 20, and the enrichment p-value is 3.25e11This is the only experimentally determined bacterial azulin protein, rubA2, rubA1, oprC, nirS, nirS, nirM, exaB, exaB, ccoQ2, ccoQ1, rubA1, azu, nirM, exaB. Responsible for annotated interactions in the STRING database. For oral cancer proteins, interactive network proteins such as CCL2, CCL3, CSF2, IL10, IL1A, IL1B, IL6, TNF, IL10 according to the gold standard set data.

Keywords: Networking, STRING, Oral cancer, Bacteria

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INTRODUCTION

Head and neck tumors are one of the 10 most common types of cancer in the world [1]. Oral squamous cell carcinoma (OSCC) in men is the most common cancer of all head and neck squamous cell carcinomas (HNSCC). It accounts for about 90% of oral malignancies and accounts for more than 300,000 newly diagnosed cases each year [2]. The number of HNSCC cases is estimated to increase to 1 million by 2030 (http://www.who.int/entity/healthinfo/statistics/bod_dalybywhoregion.xls?ua=1). Despite significant advances in cancer treatment and management, OSCC-related mortality rates have not changed. The molecular basis for aggressive OSCC growth and metastasis is still unknown. OSCCs often remain undetected at the high stages associated with high mortality and are therefore associated with high personal and social costs. The 5-year overall survival rate is estimated to be about 50% [3].Therefore, reducing oral cancer mortality requires new targets for early detection and treatment of OSCC. In recent years, identification of genes associated with complex diseases has become an important issue. Experimental approaches, such as B. genetic linkage association studies [4], expression profiling [5], and genome-wide association studies [6], are for genes at high relative risk for diseases such as cancer [7] and asthma [8]. It has been shown to be successful in identifying, Diabetes [9] and hypertension [10]. However, disease heterogeneity, the complexity of finding genes at specific loci, and the associated costs have led to the development of various in silico approaches to the identification of diseased genes.The correlation between a particular bacterium and various diseases, including cancer, is well known and well known. The role of bacteria in some types of cancer has been elucidated in more recent studies, but remains unclear in many other studies [11]. Assessing the dynamics of a microbial population under a medical condition helps determine the mechanisms by which bacteria can be used to induce or develop cancer. Periodontal disease is caused by a bacterial infection of the periodontal tissue that causes gingival inflammation and periodontal disease. Periodontitis affects the gingival tissue but not the underlying tooth support structure. Periodontal disease, in contrast, is an inflammatory disease that extends deep into the tissue and results in the loss of supporting connective tissue. Periodontitis can lead to loss of connective tissue and bone support and is the leading cause of tooth loss in adults [12]. Caries is the destruction of tooth structure by oral bacterial acids produced by microbial fermentation of leftover food [13]. Three characteristics of bacterial species are involved in biofilm formation, acid production, and caries development, including acid resistance [14] Marsh, 2005). Many acid-forming and acid-forming bacteria are involved in the carrier, including Streptococcus mutans and *Streptococcus sorbinus* (collectively known as mutans *Streptococcus*), as well as other acidic strains of lactobacillus and non-mutans streptococcus. [15]. This study follows an observational study design aimed at screening for virulence factors in the oral microflora with these proteins or oral cancer and chewing tobacco complex pathogens. This can probably interact with certain proteins. The interaction of oral cancer protein cross-linking with oral mediator proteins and other proteins was analyzed using the STRING v.5 pipeline [16] (Szklarcz). These nodes were included in the STRING database and the queries were user-defined.

MATERIAL AND METHODS

Sources and Selection of the targeting protein
The source of the plant, the geographic location of the collection, the chemical structure, and the biological activity of theoral bacterial protein azurin and oral cancer cyclin B1 protein were obtained from bibliographic sources, The origin of the plant, the geographic location of the collection, the chemical structure and biological activity of the oral bacterial proteins azurin and oral cancer cyclin B1 proteins were obtained from bibliographic sources including major journals, masters and papers, chapters of natural product chemistry. Unpublished textbooks and meeting minutes. The following standards are used by Kumar et al. [17] and Gupta et al. [18]

Protein network interaction
Extensive network analysis is used to identify functional connections between proteins and emphasize the biological importance of genes linked to enrichment pathways. The STRING and STITCH (version: 11.0) databases are global resources for predicting functional connections between proteins and cloud cluster networks and are used to study

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the interactions between proteins encoded by specific genes. Masu [19]. Experiments to identify species using P-proteins and chemicals as input gene sets. Study protein interactions (PPIs) between enriched genes and networks of interactions between them. A full score above 0.7 indicates a high level of confidence in the presence of significant interactions

RESULTS AND DISCUSSION

Protein-protein interactions are a central part of the cell network and are known to have many effects. Analyze the information flow network between all target proteins to determine the amount of information that flows between cytochrome proteins and other proteins. Use online STRING software to create a molecular genetic interaction network azurin on oral bacterial proteins, and use Cytoscape software to get 11 nodes, 16 edges, an average node order of 2.91, and visualization averages. I calculated in Table 2. The local clustering factor is 0.876, the expected number of edges is 10, and the enrichment ppi has a p-value of 0.0553. Nodes, lines, and colors show the rationality of interactive networks as shown in Figure 1 and Table 1. For interleukin 8 in oral squamous cell carcinoma, a molecular genetic interaction network was obtained and the software Cytoscape was used to visualize the number of nodes 11, the number of edges 55, the average node order 10, and the values. It was made into. The local clustering factor is 1, the expected number of edges is 20, and the enrichment p-value is ppi 3.25e11 as shown in Figure 2 and Table 3.Nodes, lines, and colors prove the rationality of interactive networks and annotation with different genes Table 4. Many experiments have shown that the gene is associated with protein expression. The results show that more residues than the cytochrome-protein cross-linking pathway are involved in protein signal intensity.

The result is represented by the color displayed in the forecast tree. A random machine learning approach [20] was used to build a reliable PPI network by combining co-movement scores obtained from metabolic partitions with detailed information suggesting physical links (Figure 2A).This is because physically interacting proteins exert similar biological activity, co-express, and often have similar evolutionary conservation. Only protein pairs with solid biochemical evidence (at least 0.4 correlation rating) from the fractionated dataset were used, and other supporting features were implemented in this subset. Two additional measurements were recorded from biochemical fractional records that reflect reproducibility. Specifically, there are a number of fractionation experiments in which the co-migration assessment of the protein pair is at least 0.4, and a number of fractionation experiments with the largest peak in each migration profile. Duplicate protein pairs. Other evidence supporting functional associations is co-[deletion] [21,22,23], interacting domains [24,25], co-evolution [26] and phenotypic records [27] (Table 3).Machine learning classifiers trained on a gold standard set containing only experimentally determined bacterial azulin proteins rubA2, rubA1, oprC, nirS, nirS, nirM, exaB, exaB, ccoQ2, ccoQ1, rubA1, azu, nirM, exaB and tested. (Table 2) is responsible for the annotated interactions in the STRING database [18]. For oral cancer proteins, interactive network proteins such as CCL2, CCL3, CSF2, IL10, IL1A, IL1B, IL6, TNF, IL10. Assessment of the relative contribution of each property to PPI prediction (measured by GiniScore) confirmed that the combined biochemical evidence had the greatest effect on classification (Table 4) and was associated with other functional associations. Compared to better co-mobility, it shows that it reflects information and is used to predict interactions.

CONCLUSION

Azurin, putative bacteriocins that have useful properties like those of azurin have been distinguished in more microscopic organisms' species. The systems of azurin and the azurin-like bacteriocins will give more and better choices in oral malignant growth treatment. In this article, we sum up how azurin and the determined peptides capture key cell controllers or cell surface receptors to redesign the phone flagging organizations. Interleukin 8 cytokines might add to the pathogenesis of this sickness, is a significant chemokine of interest in periodontal infections. The concentrate likewise reasoned that the collaboration of protein networks with different proteins can be utilized as potential remedial medication possibility for the avoidance of oral sickness.

CONFLICT OF INTEREST

None declared

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Table 1: Enrichment pathways and linked genes and nodes

Parameter	Value
Number of nodes	11
Number of edges	16
Average node degree	2.91
Avg. local clustering coefficient	0.876
Expected number of edges	10
PPI enrichment p-value	0.0553

Table 2: Networking of Azurin in oral bacterial proteinwith Annotation

Node1	Node2	Annotation node 2	Annotation node 1	Scoring function
RubA2	RubA1	Involved in the hydrocarbon hydroxylating system	which transfers electrons from NADH to rubredoxin reductase and then through rubredoxin to alkane 1 monooxygenase	0.498
RubA2	Azu	Azu transfers electrons from NADH to rubredoxin reductase and then through rubredoxin to alkane 1 monooxygenase	Transfers electrons from cytochrome c551 to cytochrome oxidase	0.685
RubA1	RubA2	Involved in the hydrocarbon hydroxylating system	It transfers electrons from NADH to rubredoxin reductase and then through rubredoxin to alkane 1 monooxygenase	0.498
RubA1	Azu	It transfers electrons from NADH to rubredoxin reductase and then through rubredoxin to alkane 1 monooxygenase	Transfers electrons from cytochrome c551 to cytochrome oxidase	0.680
OprC	Azu	Putative copper transport outer membrane porin OprC; tonB-copper: TonB-dependent copper receptor	Transfers electrons from cytochrome c551 to cytochrome oxidase	0.762
NirS	NirM	Annotation not available	Electron donor for cytochrome cd1 in nitrite and nitrate respiration	0.984
NirS	Azu	Annotation not available	Transfers electrons from cytochrome c551 to cytochrome oxidase	0.855
NirM	NirS	Electron donor for cytochrome cd1 in nitrite and nitrate respiration	annotation not available	0.984

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NirM	ExaB	Electron donor for cytochrome cd1 in nitrite and nitrate respiration	Is an essential component of the ethanol oxidation system that allows P.aeruginosa to grow on ethanol as the sole carbon and energy source.	0.691
NirM	CcoQ2	Electron donor for cytochrome cd1 in nitrite and nitrate respiration	Annotation not available	0.874
NirM	CcoQ1	Electron donor for cytochrome cd1 in nitrite and nitrate respiration	Annotation not available	0.853
NirM	Azu	Electron donor for cytochrome cd1 in nitrite and nitrate respiration	Transfers electrons from cytochrome c551 to cytochrome oxidase	0.968
ExaB	NirM	Is the direct contact between electron acceptor of the quinoprotein ethanol dehydrogenase (QEDH)	Electron donor for cytochrome cd1 in nitrite and nitrate respiration	0.691
ExaB	Azu	the direct electron acceptor of the quinoprotein ethanol dehydrogenase (QEDH)	Transfers electrons from cytochrome c551 to cytochrome oxidase	0.692
CcoQ2	NirM	Annotation not available	Electron donor for cytochrome cd1 in nitrite and nitrate respiration	0.874
CcoQ2	CcoQ1	Annotation not available	annotation not available	0.872
CcoQ2	Azu	Annotation not available	Transfers electrons from cytochrome c551 to cytochrome oxidase	0.741
CcoQ1	NirM	Annotation not available	Electron donor for cytochrome cd1 in nitrite and nitrate respiration	0.853
CcoQ1	CcoQ2	Annotation not available	annotation not available	0.872
CcoQ1	Azu	Annotation not available	Transfers electrons from cytochrome c551 to cytochrome oxidase	0.737

Table 3: Enrichment pathways and linked genes and nodes

Parameter	Value
Number of nodes	11
Number of edges	55
Average node degree	10
Avg. local clustering coefficient	1
Expected number of edges	20
PPI enrichment p-value	3.25e-11

Table 4: Networking of interleukin 8 in oral squamous cell carcinoma with Annotation

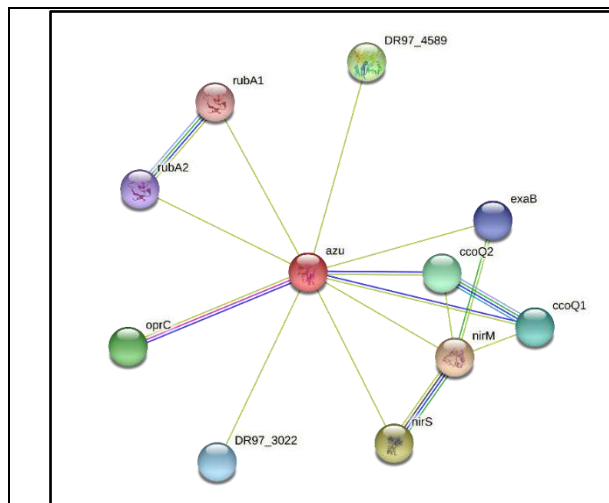| Node1	Node2	Node1 annotation	Node2 annotation	Score
CCL2	CCL3	May be involved in the recruitment of monocytes into the arterial wall during the disease process of atherosclerosis; Belongs to the intercrine beta (chemokine CC) family	Binds to CCR1, CCR4 and CCR5. One of the major HIV-suppressive factors produced by CD8+ T-cells. Recombinant MIP-1-alpha induces a dose-dependent inhibition of different strains of HIV-1, HIV-2, and simian immunodeficiency virus (SIV); Belongs to the intercrine beta (chemokine CC) family	0.921
CCL2	CSF2	Chemotactic factor that attracts monocytes and basophils but not neutrophils or eosinophils. Augments	Granulocyte-macrophage colony-stimulating factor; Cytokine that stimulates the growth and differentiation of	0.988

		monocyte anti-tumor activity. Belongs to the intercrine beta (chemokine CC) family	hematopoietic precursor cells from various lineages, including granulocytes, macrophages, eosinophils and erythrocytes; Belongs to the GM-CSF family	
CCL2	CSF3	C-C motif chemokine 2; Chemotactic factor that attracts monocytes and basophils but not neutrophils or eosinophils. Augments monocyte anti-tumor activity.	Granulocyte colony-stimulating factor; Granulocyte/macrophage colony-stimulating factors are cytokines that act in hematopoiesis by controlling the production, differentiation, and function of 2 related white cell populations of the blood, the granulocytes and the monocytes-macrophages. This CSF induces granulocytes; Belongs to the IL-6 superfamily	0.984
CCL2	CXCL2	Belongs to the intercrine beta (chemokine CC) family	Hematoregulatory chemokine, which, in vitro, suppresses hematopoietic progenitor cell proliferation. GRO-beta(5-73) shows a highly enhanced hematopoietic activity	0.984
CCL2	CXCR2	Has been implicated in the pathogenesis of diseases characterized by monocytic infiltrates, like psoriasis, rheumatoid arthritis or atherosclerosis. Belongs to the intercrine beta (chemokine CC) family	C-X-C motif chemokine 2; Produced by activated monocytes and neutrophils and expressed at sites of inflammation. Binds to IL-8 with high affinity. Also binds with high affinity to CXCL3, GRO/MGSA and NAP-2	0.929
CCL2	IL10	C-C motif chemokine 2; Chemotactic factor that attracts monocytes and basophils but not neutrophils or eosinophils. Augments May be involved in the recruitment of monocytes into the arterial wall during the disease process of atherosclerosis; Belongs to the intercrine beta (chemokine CC) family	Interleukin-10; Inhibits the synthesis of a number of cytokines, including IFN-gamma, IL-2, IL-3, TNF and GM-CSF produced by activated macrophages and by helper T-cells; Belongs to the IL-10 family	0.989
CCL2	IL1A	Augments monocyte anti-tumor activity. Has been implicated in the pathogenesis of diseases characterized by monocytic infiltrates, like psoriasis, rheumatoid arthritis or atherosclerosis.	IL-1 proteins are involved in the inflammatory response, being identified as endogenous pyrogens, and are reported to stimulate the release of prostaglandin and collagenase from synovial cells	0.983
CCL2	IL1B	Has been implicated in the pathogenesis of diseases characterized by monocytic infiltrates, like psoriasis, rheumatoid arthritis or atherosclerosis.	Promotes Th17 differentiation of T-cells	0.993
CCL2	IL6	Chemotactic factor that attracts monocytes and basophils but not neutrophils or eosinophils.	Plays an essential role in the final differentiation of B-cells into Ig- secreting cells Involved in lymphocyte and monocyte	0.994

			differentiation. Acts on B-cells, T-cells, hepatocytes, hematopoietic progenitor cells and cells of the CNS.	
CCL2	TNF	Augments monocyte anti-tumor activity.	it can stimulate cell proliferation and induce cell differentiation. Impairs regulatory T-cells (Treg) function in individuals with rheumatoid arthritis via FOXP3 dephosphorylation. Upregulates the expression of protein phosphatase 1 (PP1), which de [...]	0.992
CCL3	CCL2	Monokine with inflammatory and chemokinetic properties. Binds to CCR1, CCR4 and CCR5. One of the major HIV-suppressive factors produced by CD8+ T-cells. Recombinant MIP-1-alpha induces a dose-dependent inhibition of different strains of HIV-1, HIV-2, and simian immunodeficiency virus (SIV); Belongs to the intercrine beta (chemokine CC) family	Tumor necrosis factor; Cytokine that binds to TNFRSF1A/TNFR1 and TNFRSF1B/TNFBR. It is mainly secreted by macrophages and can induce cell death of certain tumor cell lines. It is potent pyrogen causing fever by direct action or by stimulation of interleukin-1 secretion and is implicated in the induction of cachexia, Under certain conditions	0.921
CCL3	CSF2	C-C motif chemokine 3; Monokine with inflammatory and chemokinetic properties. Binds to CCR1, CCR4 and CCR5. One of the major HIV-suppressive factors produced by CD8+ T-cells. Recombinant MIP-1-alpha induces a dose-dependent inhibition of different strains of HIV-1, HIV-2, and simian immunodeficiency virus (SIV); Belongs to the intercrine beta (chemokine CC) family	Granulocyte-macrophage colony-stimulating factor; Cytokine that stimulates the growth and differentiation of hematopoietic precursor cells from various lineages, including granulocytes, macrophages, eosinophils and erythrocytes; Belongs to the GM-CSF family	0.988
CCL3	CSF3	Monokine with inflammatory and chemokinetic properties. Binds to CCR1, CCR4 and CCR5. One of the major HIV-suppressive factors produced by CD8+ T-cells. Recombinant MIP-1-alpha induces a dose-dependent inhibition of different strains of HIV-1, HIV-2, and simian immunodeficiency virus (SIV); Belongs to the intercrine beta (chemokine CC) family	Granulocyte colony-stimulating factor; Granulocyte/macrophage colony-stimulating factors are cytokines that act in hematopoiesis by controlling the production, differentiation, and function of 2 related white cell populations of the blood, the granulocytes and the monocytes-macrophages. This CSF induces granulocytes; Belongs to the IL-6 superfamily	0.986
CCL3	CXCL2	C-C motif chemokine 3; Monokine with inflammatory and chemokinetic properties. Binds to CCR1, CCR4 and CCR5. One of the major HIV-suppressive factors produced by	C-X-C motif chemokine 2; Produced by activated monocytes and neutrophils and expressed at sites of inflammation. Hematoregulatory chemokine, which, in vitro, suppresses hematopoietic progenitor	0.983

		CD8+ T-cells. Recombinant MIP-1-alpha induces a dose-dependent inhibition of different strains of HIV-1, HIV-2, and simian immunodeficiency virus (SIV); Belongs to the intercrine beta (chemokine CC) family	cell proliferation. GRO-beta(5-73) shows a highly enhanced hematopoietic activity	
CCL3	CXCR2	Binds to CCR1, CCR4 and CCR5. One of the major HIV-suppressive factors produced by CD8+ T-cells. Recombinant MIP-1-alpha induces a dose-dependent inhibition of different strains of HIV-1, HIV-2, and simian immunodeficiency virus (SIV); Belongs to the intercrine beta (chemokine CC) family	C-X-C chemokine receptor type 2; Receptor for interleukin-8 which is a powerful neutrophil chemotactic factor. Binding of IL-8 to the receptor causes activation of neutrophils. This response is mediated via a G-protein that activates a phosphatidylinositol-calcium second messenger system. Binds to IL-8 with high affinity. Also binds with high affinity to CXCL3, GRO/MGSA and NAP-2	0.899
CCL3	IL10	C-C motif chemokine 3; Monokine with inflammatory and chemokinetic properties. Binds to CCR1, CCR4 and CCR5. One of the major HIV-suppressive factors produced by CD8+ T-cells. Recombinant MIP-1-alpha induces a dose-dependent inhibition of different strains of HIV-1, HIV-2, and simian immunodeficiency virus (SIV); Belongs to the intercrine beta (chemokine CC) family	Interleukin-10; Inhibits the synthesis of a number of cytokines, including IFN-gamma, IL-2, IL-3, TNF and GM-CSF produced by activated macrophages and by helper T-cells; Belongs to the IL-10 family	0.988
CCL3	IL1A	One of the major HIV-suppressive factors produced by CD8+ T-cells.	Interleukin-1 alpha; Produced by activated macrophages, IL-1 stimulates thymocyte proliferation by inducing IL-2 release, B-cell maturation and proliferation, and fibroblast growth factor activity. IL-1 proteins are involved in the inflammatory response, being identified as endogenous pyrogens, and are reported to stimulate the release of prostaglandin and collagenase from synovial cells	0.984
CCL3	IL1B	Recombinant MIP-1-alpha induces a dose-dependent inhibition of different strains of HIV-1, HIV-2, and simian immunodeficiency virus (SIV); Belongs to the intercrine beta (chemokine CC) family	Interleukin-1 beta; Potent proinflammatory cytokine. Initially discovered as the major endogenous pyrogen, induces prostaglandin synthesis, neutrophil influx and activation, T-cell activation and cytokine production, B-cell activation and antibody production, and fibroblast proliferation and collagen production. Promotes Th17 differentiation of T-cells	0.996
CCL3	IL6	C-C motif chemokine 3; Monokine	It is a potent inducer of the acute phase	0.989

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		with inflammatory and chemokinetic properties. Binds to CCR1, CCR4 and CCR5. One of the major HIV-suppressive factors produced by CD8+ T-cells. Recombinant MIP-1-alpha induces a dose-dependent inhibition of different strains of HIV-1, HIV-2, and simian immunodeficiency virus (SIV); Belongs to the intercrine beta (chemokine CC) family	response. Plays an essential role in the final differentiation of B-cells into Ig- secreting cells Involved in lymphocyte and monocyte differentiation. Acts on B-cells, T-cells, hepatocytes, hematopoietic progenitor cells and cells of the CNS. Required for the generation of T(H)17 cells. Also acts as a myokine.	
CCL3	TNF	C-C motif chemokine 3; Monokine with inflammatory and chemokinetic properties. Binds to CCR1, CCR4 and CCR5. One of the major HIV-suppressive factors produced by CD8+ T-cells. Recombinant MIP-1-alpha induces a dose-dependent inhibition of different strains of HIV-1, HIV-2, and simian immunodeficiency virus (SIV); Belongs to the intercrine beta (chemokine CC) family	Tumor necrosis factor; Cytokine that binds to TNFRSF1A/TNFR1 and TNFRSF1B/TNFBR.. Impairs regulatory T-cells (Treg) function in individuals with rheumatoid arthritis via FOXP3 dephosphorylation. Upregulates the expression of protein phosphatase 1 (PP1),	0.993

Role of interleukin 8 in oral squamous cell carcinoma

Figure 1.Azurin protein network nodes interaction	**Figure 2.Interleukin 8protein network nodes interaction**

RESEARCH ARTICLE

Evaluation of Antibacterial and Antifungal Activity of Selected Fruit Peels of Family Cucurbitaceae.

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ABSTRACT

Presence of different phytochemicals are responsible for therapeutic properties of a plant. In recent years, different plant-based drugs are being discovered as antimicrobial agents due to development of resistance to traditional antibiotic drugs. In the present investigation, methanolic extracts of fruit peels of some members of family Cucurbitaceae (*Citrullus lanatus, Cucumis sativus, Lagenaria siceraria, Momordica charantia, Cucumis melo, and Cucurbita maxima*) were evaluated for their antibacterial (against *E. coli*, and *S. aureus*), and antifungal (*A. niger*, and *P. chrysogenum*) at different concentrations (30 mg/L, 60 mg/L, and 90 mg/L) along with standard antibiotic drug (as positive control) by disc diffusion assay. Inhibition Zone (IZ), Activity Index (AI), and Minimum Inhibitory Concentration (MIC) were calculated for each extract. Results showed the presence of good activity of the extracts against the selected microbes. Against *E. coli*, maximum antibacterial activity was shown by peel extract of *Cucumis sativus* with the lowest MIC (20-25 mg/L) while against *S. aureus*, maximum inhibitory activity was observed by peel extract of *C. sativus* and *C. melo* with the lowest MIC value (20-25 mg/ml). The lowest MIC (10-15) was shown by *C. sativus, L. siceraria, M. charantia* (10-15 mg/L) against *A. niger* while against *P. chrysogenum*, the maximum activity was shown by *L. siceraria* and *C. melo* (25-30 mg/ml). Results of the present study indicated that these fruit peels, which are thrown as waste materials, can be utilized as rich sources of antimicrobial agents and by isolation, and identification of responsible phytochemicals, research may be helpful for pharmaceutical industries to formulate antimicrobial drug.

Keywords: fruit peels, cucurbits, antibacterial, antifungal, MIC etc.

INTRODUCTION

An increase in microbial resistance to antimicrobial agents has been worldwide during past decades (Livermore, D.M., 1995; Jones, R.N.*et al.*, 1997; Kolar, M.*et al.*, 2001). Nowadays, it is critically needed to discover new antibacterial and antifungal drugs against different infectious diseases because many deaths occur due to these diseases every year throughout the world and to the existed antimicrobial drugs, resistance has been developed into pathogens. Green plants represent a useful source of reservoir of effective chemotherapeutics and can provide valuable source of natural antimicrobials (Balandrin, M.F.*et al.*, 1985; Satish, S.*et al.*, 1999). Antimicrobials of plant origin are effective in the treatment of infectious diseases while fewer side effects often associated with natural products as compared to synthetic antimicrobials (Kokosha, L.*et al.*, 2002). Cucurbits, belongs to the family Cucurbitaceae, are the most consumed food cropthroughout worlds. Cucurbits are excellent fruits in nature, having a composition of all the essential constituents required for the good health of humans (Rahman, A.S., 2003). Consumption of bottle gourd has been associated with a number of health benefits and can be regarded as a natural protector against diseases (Barot, A.*et al.*, 2015). The industrial waste which is generated in large amount can be processed to use as fertilizers but generally, kitchen waste like peels of fruits and vegetables are thrown in the garbage.

Various studies confirmed that kitchen waste materials like peels of fruits and vegetables can be successfully processed and further utilized as sources of medicinal compounds. Studies have revealed the presence of a wide range of secondary metabolites, including flavonoids, triterpenoid, saponins, and phenolic acids in gourd vegetables possessing distinct biological activities (Rizvi, M.M.A., 2009). In the present investigation, fruit peels of some cucurbits (*Citrullus lanatus, Cucumis sativus, Lagenaria siceraria, Momordica charantia, Cucumis melo, and Cucurbita maxima*) were evaluated for their antibacterial and antifungal activity.

MATERIALS AND METHODS

Collection Of Plants And Extract Preparation
Fresh fruit samples were purchased from local market of Jaipur, Rajasthan. Those were taken to the laboratory to be washed and dried in air at room temperature. Peels were separated from the samples. For determination of moisture content, all the collected peels were kept in a flat-bottom dish in an air oven at 105°C for 1,3,5 hours and allowed to cool at room temperature in a desiccator and then weighed. The procedure was repeated until successive weighing agrees to as constant weighing. The loss in weight of the plant material was regarded as a measure of moisture content. The air-dried and coarsely powered, approximately 30 g of plant samples were places in Soxhlet extractor with 150 ml methanol for 24 h. Then, these were filtered using Whatman filter paper No. 1. The moisture were evaporated and the extracts were dried using rotary evaporator under controlled temperature and low pressure. The obtained extracts were dried over anhydrous $CaCl_2$ and utilized for further evaluation.

Antimicrobial Analysis
Dried plant extracts were used for evaluate their antimicrobial activity. These were investigated by using well diffusion assay method. For this, 2 bacterial and 2 fungal strains were used to investigate antimicrobial activities. Pure cultures of bacterial isolates of *Staphylococcus aureus* MTCC 87, *Escherichia coli* MTCC 1652, and pure culture of fungal isolates *viz. Aspergillus niger* NCIM (National Collection of Industrial Microorganisms, Pune) 0616, *Penicillium chrysogenum* MTCC 2725, were collected from Department of Botany, University of Rajasthan, Jaipur, India, which were used as indicator organisms. The fungal cultures were sub-cultured on sterile Potato Dextrose Agar (PDA) and incubated at 28°C for 48 hrs. The bacteria were grown in Nutrient Agar (NA) medium (prepared by autoclaving 8% Nutrient agar in distilled water at 15lbs psi for 25-30 min) and incubating at 37°C for 48 hrs. All fungal and bacterial cultures were further maintained on the same medium after every 48 hrs. Fresh suspensions of test organisms in saline solution were prepared from freshly grown agar slants before every antimicrobial assay.

Antibacterial Assay

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Standard microbial technique- the Agar Well Diffusion method (Perez, C. *et al.*, 1990)was used for *in-vitro* antibacterial assay. The different samples were diluted by using 10% di-methyl sulphoxide (DMSO) and 3 different concentrations (30 mg/L, 60 mg/L, and 90 mg/L) of all extracts were prepared. Disinfected Petri dishes holding the nutrient agar (NA) medium were inoculated with the microorganisms under investigation with spreader and allowed to stand for 30 min. Wells of 6mm diameter were prepared in the seeded agar plates. With equal distance control well was also prepared. All different concentrations of all the samples and standard drug (30μl) poured into the preorganized wells of seeded plates. Then plates were kept for incubation for 24 hrs at 37°C. The antibacterial spectrum of the test sample was determined in terms of inhibition zone (IZ) around each well. The diameters of inhibition zone produced by the test sample were compared with the inhibition zone, produced by the commercial control antibiotics ciprofloxacin (1mg/ml).

Antifungal Assay

Antifungal activity was screened by modified agar well diffusion method (Bonjar, G.H.S., 2004). Petri plates having PDA medium were inoculated with the fungal strains (7 days old) separately suspended in saline solution. The plates were dried out at room temperature for 15 min. Wells of 6 mm in diameter were perforated on the agar using cork-borers. With equal distance control well was also prepared. The different samples were diluted by using 10% di-methyl sulphoxide (DMSO) and 3 different concentrations (30 mg/L, 60 mg/L, and 90 mg/L) of all the extracts, were also prepared. All different concentrations of all samples and standard drug seeded into the wells of preorganized Patri plates. Seeded plates were incubated at 28°C for the 48 hours. After incubation, antifungal activities were determined by computing the diameter of IZ (mm). All experimentations were made thrice and mean values were calculated. Ketokenazole (1mg/ml) was used as the standard control for antifungal assay.

Determination of Activity Index (AI)

The activity index (AI) for all the plant extracts were calculated as:

$$\text{Activity index (A.I.)} = \frac{\text{Mean of inhibition zone of the extract}}{\text{inhibition zone obtained for standard antibiotic drug}}$$

Determination of Minimum Inhibitory Concentration (MIC)

The Macro-broth dilution technique was used to determine minimum inhibitory concentration of the extracts by diluting a given volume of the extract to various concentrations (Baron, E.J.*et al.*, 1990). Wells of 6 mm were prepared on agar surface using sterile cork bore and these were labelled properly. The different concentrations were used to fill up the wells using Pasteur pipette. This was followed by the incubation of the plates at 37°C for 24 hours for bacteria and 28°C for 48 hours for fungi. The minimum dilution of sample that inhibits the growth of the organism was taken as Minimum Inhibitory Concentration (Mathur, A.*et al.*, 2011).

RESULTS AND DISCUSSION

Various studies have shown that different infectious agents causing human diseases have been developed resistant against known antimicrobial drugs. It has also been confirmed that existing antimicrobial drugs impact health negatively also. Therefore, it is important to survey for new proxies that are better, less expensive and without side effect treating infectious diseases, especially in developing countries. Natural products obtained from plants are being utilized for treatment of diseases at large scale. Phytoconstituents have been found to showing a promising effect on microorganisms, parasites, viruses and pests (Garcia-Olmedo, F. *et al.*, 1998). In the present study, methanolic extracts of fruit peels of 6 different plants (*Citrullus lanatus, Cucumis sativus, Lagenaria siceraria, Momordica charantia, Cucumis melo, and Cucurbita maxima*) of family Cucurbitaceae were subjected for their antibacterial (*E. coli, and S. aureus*) and antifungal (*A. niger, and P. chrysogenum*) activity. Ciproflaxin was used as standard (positive control). Three concentrations (30 mg/L, 60 mg/L, and 90 mg/L) of each extract were evaluated against all microbes. Results of the present investigation revealed that all the selected fruit peel extracts have good impact against the

selected microorganisms (Table 1). Inhibition zone (IZ), activity index (AI), and minimum inhibitory concentration (MIC) for all extracts against all microorganism were calculated separately. Against *E. coli*, maximum antibacterial activity was shown by peel extract of *Cucumis sativus* with the lowest MIC (20-25 mg/L) followed by *Lagenaria siceraria* (25-30 mg/L), *C. sativus*, *C. melo* (35-40 mg/L) while extracts of peels of *C. maxima* and *C. lanatus* showed higher MIC values (110-120 mg/L and 125-130 mg/L respectively) as compared to others (Figure 1). Against *S. aureus*, maximum inhibitory activity was observed by peel extract of *C. sativus* and *C. melo* with the lowest MIC value (20-25 mg/ml), MIC values of other extracts were not significantly different from this value except *C. lanatus* (80-85 mg/L) (Figure 2).

Extracts of selected fruit peels showed good activity against *A. niger*. The lowest MIC (10-15) was shown by *C. sativus*, *L. siceraria, M. charantia* (10-15 mg/L). other MIC values were not significantly different(Figure 3) while against *P. chrysogenum*, maximum activity was shown by *L. siceraria* and *C. melo* (25-30 mg/ml) (Figure 4). It has been previously investigated by many researchers and scientists that phytochemicals can be utilized as antimicrobial compounds and can be helpful to to control growth of food borne and spoilage bacteria. Some researchers have suggested that antimicrobial components of the plant extracts (terpenoid, alkaloid and phenolic compounds) interact with enzymes and proteins of the microbial cell membrane causing its disruption to disperse a flux of protons towards cell exterior which induces cell death or may inhibit enzymes necessary for aminoacids biosynthesis (Burt, S., 2004, Gill and Holley, 2006).In our previous studies, we observed the presence of different phytochemicals in the selected fruit peels (Khatana, S. *et al.*, 2021). Other researchers attributed the inhibitory effect of these plant extracts to hydrophobicity characters of these plants extracts which enable them to react with protein of microbial cell membrane and mitochondria disturbing their structures and changing their permeability (Friedman,*et al.*, 2004, Tiwari,*et al.*, 2009). The present study suggested that plant parts which are thrown as waste material can be used as natural source of antimicrobial agents.

CONCLUSION

The results of the present study have concluded that, fruit peels of the selected cucurbit plants are rich source of antimicrobial agents. So, these can be further utilized for formulation of therapeutic drugs for infectious diseases alone or in combinatorial therapy.However, Isolation, identification and purification of responsible phytoconstituents and determination of their respective antimicrobial potencies and toxicological evaluation with the view of formulating novel chemotherapeutic agents should be the future path of investigation.

ACKNOWLEDGEMENT

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Table 1: Antimicrobial activity of methanolic extracts of different fruit peels of the selected cucurbits.

Name of the plant	Standard	30 mg/L		60 mg/L		90 mg/L		MIC (mg/L)
E. coli	IZ	IZ	AI	IZ	AI	IZ	AI	
Citrullus lanatus	41	NA	NA	NA	NA	NA	NA	125-130
Cucumis sativus	43	13	0.30	16	0.37	20	0.46	20-25
Lagenaria siceraria	46	14	0.30	17	0.36	21	0.45	25-30
Momordica charantia	51	11	0.21	12	0.23	14	0.27	35-40
Cucumis melo	41	11	0.26	12	0.29	15	0.36	35-40
Cucurbita maxima	40	NA	NA	NA	NA	NA	NA	110-120
S. aureus								
Citrullus lanatus	51	NA	NA	NA	NA	11	0.21	80-85
Cucumis sativus	50	11	0.22	14	0.28	15	0.30	20-25
Lagenaria siceraria	46	NA	NA	12	0.26	16	0.34	40-45
Momordica charantia	51	NA	NA	12	0.23	14	0.27	45-50
Cucumis melo	45	9	0.20	12	0.26	14	0.31	20-25

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Cucurbita maxima	48	10	0.20	12	0.25	14	0.29	25-30
A. niger								
Citrullus lanatus	35	12	0.34	21	0.60	24	0.68	15-20
Cucumis sativus	30	13	0.43	21	0.70	27	0.90	10-15
Lagenaria siceraria	38	12	0.31	20	0.52	22	0.57	10-15
Momordica charantia	36	15	0.41	20	0.55	26	0.72	10-15
Cucumis melo	34	12	0.35	21	0.61	30	0.88	15-20
Cucurbita maxima	30	15	0.50	21	0.70	27	0.90	10-15
P. chrysogenum								
Citrullus lanatus	27	NA	NA	11	0.40	16	0.59	50-55
Cucumis sativus	28	NA	NA	9	0.32	11	0.39	55-60
Lagenaria siceraria	30	9	0.30	13	0.43	14	0.46	25-30
Momordica charantia	26	9	0.34	11	0.42	16	0.61	50-55
Cucumis melo	27	9	0.33	13	0.48	14	0.51	25-30
Cucurbita maxima	32	NA	NA	NA	NA	11	0.34	75-80

Note: IZ- Inhibition Zone (mm), AI- Activity Index, MIC- Minimum Inhibitory Concentration (mg/L), S- Standard antimicrobial drug (positive control), NA- No Activity
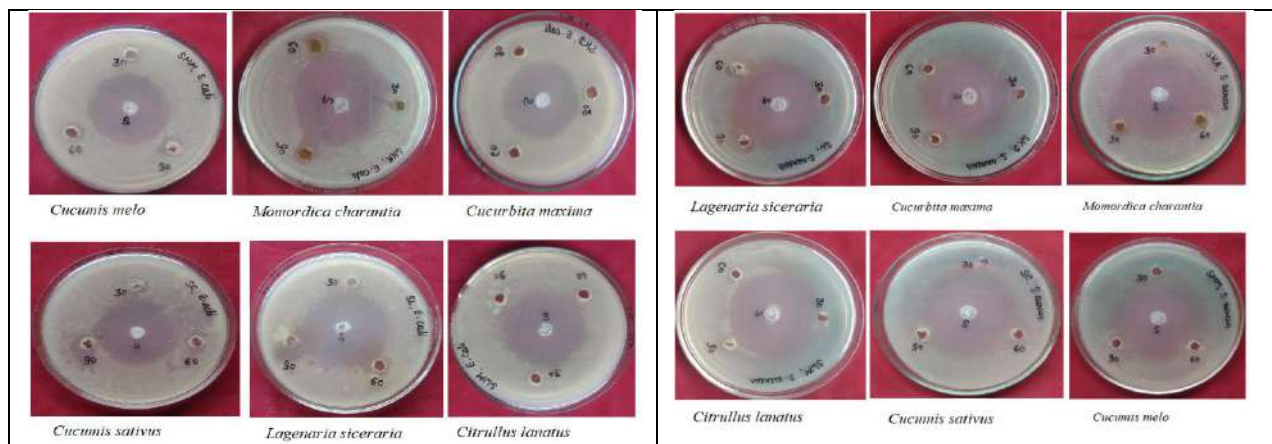
Figure 1: Antibacterial activity of methanolic extracts of fruit peels of different cucurbits against *E. coli*.

Figure 2: Antibacterial activity of methanolic extracts of fruit peels of different cucurbits against *S. aureus*.
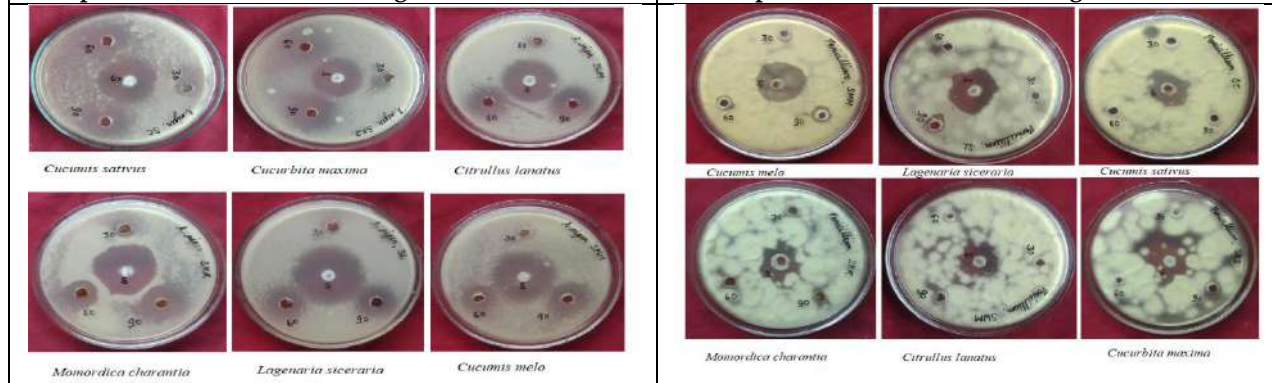
Figure 3:Antifungalactivity of methanolic extracts of fruit peels of different cucurbits against *A. niger*.

Figure 4: Antifungal activity of methanolic extracts of fruit peels of different cucurbits against *P. chrysogenum*

RESEARCH ARTICLE

Fuzzy EOQ Model for Deteriorating Products Using Preservation Technology

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ABSTRACT

The paper studies a kind of deteriorating seasonal product whose deterioration rate can be controlled by investing on the preservation efforts. Inventory control is influenced by factors like demand, holding costs, deterioration costs, lost sales, and shortage costs, etc. Since these characteristics are vague in real-world problems, the current study developed a fuzzy economic order quantity (EOQ) inventory model with a partial backlog. Preservation technology is being used to slow down the rate of deterioration. Parameters are taken as triangular fuzzy numbers. The model is developed to find fuzzy total average cost function which is defuzzified by using the Signed distance method (SDM) and Centroid method (CM) and the results are compared. A numerical example has been given in crisp as well as in fuzzy approach to demonstrate and validate the proposed model. Finally, the effects of different parameters have been rigorously studied through sensitivity analysis.

Keywords: Inventory, deterioration, triangular fuzzy number, Signed distance method, Centroid method

INTRODUCTION

Inventory is a physical stock that a business owner maintains in mind in order to ensure that his or her operations function smoothly and efficiently. Developing countries like India, Pakistan, Sri Lanka, Bhutan, Thailand, and Bangladesh, who rely solely on their own agriculture, are facing grave harvesting scenarios during this global

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pandemic. In such circumstances, supplying decaying products is a nightmare and extremely costly. As a result, there is a significant economic loss. Small cities in East and South India, such as Odisha, West Bengal, Jharkhand, Tamil Nadu, Kerala, and Karnataka, have found that vegetable items are sometimes accessible at a very low price and sometimes not at all due to a lack of cold storage. In Odisha, the price of vegetables such as tomatoes, onions, and cauliflower is so low during the harvesting season that farmers have been known to leave the crop in the field to be destroyed. As a result, they are confronted with financial difficulties, prompting them to consider suicide. In other non-seasonal months of the year, however, these vegetables are extremely expensive. We developed a model of decaying products utilizing preservation technology to account for these issues. We assumed that the above items could be obtained from local farmers, preserved using preservation technology, and then sold at their highest value. Finally, the farmers will pay their loan amount. In this model we use fuzzy technology since the economic parameters like holding cost, demand, deterioration cost, lost sale cost, shortage cost probably will have some little fluctuations or vague in nature. So in these practical situations, we treat these parameters as triangular fuzzy variables which will be more realistic. Hence fuzzy set theory is necessary for the formulation of such inventory models. In 1965, Zadeh the first person introduced the concept of fuzziness. Later, Zadeh and Bellman (1970) applied fuzzy set theory on the development of an inventory model. Thus, this new development of fuzzy concept gives more accurate result as compared to classical probability theory. Many researchers attracted towards the development of inventory models applying fuzzy concept. It is found that fuzzy inventory models have been developed by Roy and Maiti (1998), Maiti and Maiti (2006), Rong, Mahaptra and Maiti (2008), Roy *et al.* (2009), Yadav *et al.* (2012), Kumar *et al.* (2013), Singh and Anuradha (2014), Raula *et al.* (2018), Indrajitsingha *et al.* (2018, 2019, 2020) and many others. Most of them consider the fuzzy variables as triangular fuzzy number and for defuzzification used graded mean integration representation method, signed distance method or centroid method.

Deterioration plays an important role while developing inventory models of seasonal products like potatoes, onions, food-grains etc. To slow down the deterioration, preservation technology cost may be used for the preservation of these above products, so that one can get more profit as compared to previous models. During the last decade of the present century, use of preservation technology cost while developing inventory model , in various times, had been considered by different scientist such as Hsu *et al.* (2010), Dye and Hsieh (2012), Dye (2013), Yang *et al.* (2015), Singh *et al.* (2016), Saha *et al.* (2017), pal *et al.* (2018), Mishra *et al.* (2018), Mishra *et al.* (2019), Das *et al.* (2020).

In the present paper, we develop an inventory framework of deteriorating items applying preservation technologies to minimize the deterioration rate in which the preservation technology investment parameter and critical point are assumed to be the decision variables.

Model Notation And Assumptions
Notations
Throughout the paper we use the following notations:

$d(t)$	:	Demand rate.
α	:	Initial demand rate.
β	:	Positive demand coefficient.
$m(\xi)$	:	Deterioration rate, after applying preservation technology.
m_o	:	The deterioration rate, before applying preservation technology.
δ	:	Sensitive parameter of investment to the deterioration rate.
h	:	Inventory holding cost per unit per unit time.
s	:	Shortage cost per unit.
l	:	Lost sale cost per unit.
c	:	Deterioration cost per unit.
t_1	:	Time at which inventory level becomes zero.
A	:	Ordering cost per order.
Q	:	Initial inventory level.
T	:	The duration of the replenishment cycle .

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k	:	A constant, $0 < k \le 1$.
$\tilde{\alpha}$	:	Fuzzy initial demand rate.
$\tilde{\beta}$	:	Fuzzy positive demand coefficient.
$\tilde{h}$	:	Fuzzy inventory holding cost per unit per unit time.
$\tilde{c}$	:	Fuzzy deterioration cost per unit.
$\tilde{l}$	:	Fuzzy lost sale cost per unit.
$\tilde{s}$	:	Fuzzy shortage cost.
$TAC(\xi, t_1)$		Total average cost, $0 \le t_1 \le T$.
$\widetilde{TAC}(\xi, t_1)$		Fuzzy total average cost.
$\widetilde{TAC}_g(\xi, t_1)$		Defuzzified value of $\widetilde{TAC}(\xi, t_1)$ applying GMIR method, $0 \le t_1 \le T$.
$\widetilde{TAC}_s(\xi, t_1)$		Defuzzified value of $\widetilde{TAC}(\xi, t_1)$ applying Signed distance method, $0 \le t_1 \le T$.
$\widetilde{TAC}_c(\xi, t_1)$		Defuzzified value of $\widetilde{TAC}(\xi, t_1)$ applying Centroid method, $0 \le t_1 \le T$.

Assumptions

The model is established by considering the following assumptions:

i.　　　Demand rate is assumed to be $d(t) = \alpha + \beta I_1(t)$

ii.　　The inventory system involves only one product.

iii.　　The market demand depends only on the availability of stock.

iv.　　The lead time is assumed to be negligible.

v.　　Shortages are allowed with partially backlogging.

vi.　　There is no cost to dispose or store deteriorated products.

vii.　　It is assumed that $m(\xi) = m_o e^{-\delta \xi}$.

Mathematical Model Development

Suppose the system is considered with initial stock Q units and at time $t = t_1$, the inventory level reduces to zero due to the combined effect of deterioration and demand. After that shortage occurs in the system it is backlogged partially. In the present article a single retailer's inventory model is considered in which deteriorating rate is considered by using preservation technology. Using the above assumptions, the inventory level pattern is depicted in fig. 1.

Thus, the inventory system can be described by the following differential equations:

$$\frac{dI_1(t)}{dt} + m_o e^{-\delta \xi} I_1(t) = -d(t), \ 0 \le t \le t_1 \tag{1}$$

$$\frac{dI_2(t)}{dt} = -k\xi, \ t_1 \le t \le T \tag{2}$$

with boundary conditions

$$I_1(0) = Q, \ I_1(t_1) = 0 \tag{3}$$

The solutions of these equations are given by

$$I_1(t) = \frac{\alpha}{\beta + m_o e^{-\delta \xi}}\left[e^{(\beta + m_o e^{-\delta \xi})(t_1 - t)} - 1\right] \tag{4}$$

$$I_2(t) = k\xi(t_1 - t) \tag{5}$$

Using equations (5) and (6) the value of the parameters has been calculated:

Economic order quantity

$$Q = \frac{\alpha}{\beta + m_o e^{-\delta \xi}}\left[e^{(\beta + m_o e^{-\delta \xi})t_1} - 1\right] \tag{6}$$

Holding cost

$$HC = h\left\{\frac{\alpha}{\beta}\left(\beta + m_o e^{-\delta \xi}\right)\right\}\frac{t_1^2}{2} \tag{7}$$

Deterioration cost

$$DC = c\left[\frac{\alpha}{\beta}\left(e^{(\beta + m_o e^{-\delta \xi})} - 1\right) - \left(\alpha t_1 + \alpha\beta\frac{t_1^2}{2}\right)\right] \tag{8}$$

Ordering cost

$$OC = A \tag{9}$$

Shortage cost

$SC = ks\xi(T - t_1)$ (10)

Lost sale cost

$LSC = l\xi(1 - k)(T - t_1)$ (11)

Preservation technology cost

$PTC = \xi T$ (12)

The average total cost $TAC(\xi, t_1)$ for the period T

$TAC(\xi, t_1) = \frac{1}{T}[HC + DC + OC + SC + LSC + PTC]$

$$= \frac{1}{T}\left[h\left\{ \frac{\alpha}{\beta}(\beta + m_o e^{-\delta\xi}) \right\}\frac{t_1^2}{2} + c\left\{ \frac{\alpha}{\beta}\left(e^{(\beta + m_o e^{-\delta\xi})} - 1 \right) - \left(\alpha t_1 + \alpha\beta\frac{t_1^2}{2} \right) \right\} + A \atop + ks\xi(T - t_1) + l\xi(1 - k)(T - t_1) + \xi T \right]$$ (13)

To minimize the average total cost function $TAC(\xi, t_1)$ per unit time, the value of t_1 and ξ that minimizes average cost can be obtained by solving the equations

$\frac{\partial ATC(\alpha, t_1)}{\partial \xi} = 0$ and $\frac{\partial ATC(\alpha, t_1)}{\partial t_1} = 0$ (14)

satisfying the conditions

$\frac{\partial^2 TAC(\xi, t_1)}{\partial \xi^2} > 0, \frac{\partial^2 TAC(\xi, t_1)}{\partial t_1^2} > 0$ and

$\left(\frac{\partial^2 TAC(\xi, t_1)}{\partial \xi^2} \right)\left(\frac{\partial^2 TAC(\xi, t_1)}{\partial t_1^2} \right) - \left(\frac{\partial^2 TAC(\xi, t_1)}{\partial \xi \partial t_1} \right)^2 > 0$ (15)

Equations (14) are equivalent to

$\frac{1}{T}\left[T - \frac{\alpha}{\beta}\left(m_o c e^{\beta - \delta\xi + m_o e^{-\delta\xi}} \right) - \frac{\alpha h \delta t_1^2 m_o e^{-\delta\xi}}{2\beta} \right] = 0$

and

$\frac{1}{T}\left[\frac{\alpha}{\beta} h t_1(\beta + m_o e^{-\delta\xi}) - \alpha c(1 + \beta t_1) - \alpha l(1 - k) - \alpha ks \right] = 0$

Fuzzy Model

Due to uncertainty in nature, it is not easy to define all the system parameters exactly. Hence, it is assumed that some of the parameters namely $\tilde{\alpha}, \tilde{\beta}, \tilde{h}, \tilde{c}, \tilde{l}$ may change within some limits. Suppose $\tilde{\alpha} = (\alpha_1, \alpha_2, \alpha_3)$, $\tilde{\beta} = (\beta_1, \beta_2, \beta_3)$, $\tilde{h} = (h_1, h_2, h_3)$, $\tilde{c} = (c_1, c_2, c_3)$, $\tilde{s} = (s_1, s_2, s_3)$ and $\tilde{l} = (l_1, l_2, l_3)$ are considered as triangular fuzzy numbers. Then the fuzzy total average cost $\widehat{TAC}(\xi, t_1)$ is given by

$$\widehat{TAC}(\xi, t_1) = \frac{1}{T}\left[\tilde{h}\left\{ \frac{\tilde{\alpha}}{\tilde{\beta}}(\tilde{\beta} + m_o e^{-\delta\xi}) \right\}\frac{t_1^2}{2} + \tilde{c}\left\{ \frac{\tilde{\alpha}}{\tilde{\beta}}\left(e^{(\tilde{\beta} + m_o e^{-\delta\xi})} - 1 \right) - \left(\tilde{\alpha}t_1 + \tilde{\alpha}\tilde{\beta}\frac{t_1^2}{2} \right) \right\} + A \atop + k\tilde{s}\xi(T - t_1) + \tilde{l}\xi(1 - k)(T - t_1) + \xi T \right]$$ (16)

Defuzzifying the fuzzy total average cost $\widehat{TAC}(\xi, t_1)$ by the following three different methods, we get

(i)   Signed distance method

$\widehat{TAC}_s(\xi, t_1) = \frac{1}{4}\left[\widehat{TAC}_{s1}(\xi, t_1) + 2\widehat{TAC}_{s2}(\xi, t_1) + \widehat{TAC}_{s3}(\xi, t_1) \right]$ (17)

where

$$\widehat{TAC}_{si}(\xi, t_1) = \frac{1}{T}\left[h_i\left\{ \frac{\alpha_i}{\beta_i}(\beta_i + m_o e^{-\delta\xi}) \right\}\frac{t_1^2}{2} + c_i\left\{ \frac{\alpha_i}{\beta_i}\left(e^{(\beta_i + m_o e^{-\delta\xi})} - 1 \right) - \left(\alpha_i t_1 + \alpha_i\beta_i\frac{t_1^2}{2} \right) \right\} + A \atop + ks_i\xi(T - t_1) + l_i\xi(1 - k)(T - t_1) + \xi T \right],$$

$$for\ i = 1,2,3$$

To minimize the total average cost function $\widehat{TAC}_s(\xi, t_1)$ per unit time, the optimal value of t_1 and ξ can be obtained by solving the equations

$\frac{\partial \widehat{TAC}_s(\xi, t_1)}{\partial \xi} = 0$ and $\frac{\partial \widehat{TAC}_s(\xi, t_1)}{\partial t_1} = 0$ (18)

satisfying

$\frac{\partial^2 \widehat{TAC}_s(\xi, t_1)}{\partial \xi^2} > 0, \frac{\partial^2 \widehat{TAC}_s(\xi, t_1)}{\partial t_1^2} > 0$ and

$\left(\frac{\partial^2 \widehat{TAC}_s(\xi, t_1)}{\partial \xi^2} \right)\left(\frac{\partial^2 \widehat{TAC}_s(\xi, t_1)}{\partial t_1^2} \right) - \left(\frac{\partial^2 \widehat{TAC}_s(\xi, t_1)}{\partial \xi \partial t_1} \right)^2 > 0$ (19)

(ii)   Centroid method

$$\widetilde{TAC_c}(\xi, t_1) = \frac{1}{3}\left[\widetilde{TAC_c}_1(\xi, t_1) + \widetilde{TAC_{c2}}(\xi, t_1) + \widetilde{TAC_{c3}}(\xi, t_1)\right] \tag{20}$$

where

$$\widetilde{TAC_{ci}}(\xi, t_1) = \frac{1}{T}\left[h_i\left\{\frac{\alpha_i}{\beta_i}(\beta_i + m_o e^{-\delta\xi})\right\}\frac{t_1^2}{2} + c_i\left\{\frac{\alpha_i}{\beta_i}\left(e^{(\beta_i + m_o e^{-\delta\xi})} - 1\right) - \left(\alpha_i t_1 + \alpha_i\beta_i\frac{t_1^2}{2}\right)\right\} + A, \atop +ks_i\xi(T - t_1) + l_i\xi(1 - k)(T - t_1) + \xi T \right]$$
$$for\ i = 1,2,3$$

To minimize the total average cost function $\widetilde{TAC_c}(\xi, t_1)$ per unit time, the optimal value of t_1 and ξ can be obtained by solving the equations

$$\frac{\partial \widetilde{TAC_c}(\xi, t_1)}{\partial \xi} = 0 \quad \text{and} \quad \frac{\partial \widetilde{TAC_c}(\xi, t_1)}{\partial t_1} = 0 \tag{21}$$

satisfying the equations

$$\frac{\partial^2 \widetilde{TAC_c}(\xi, t_1)}{\partial \xi^2} > 0, \ \frac{\partial^2 \widetilde{TAC_c}(\xi, t_1)}{\partial t_1^2} > 0 \ \text{ and }$$

$$\left(\frac{\partial^2 \widetilde{TAC_c}(\xi, t_1)}{\partial \xi^2}\right)\left(\frac{\partial^2 \widetilde{TAC_c}(\xi, t_1)}{\partial t_1^2}\right) - \left(\frac{\partial^2 \widetilde{TAC_c}(\xi, t_1)}{\partial \xi \partial t_1}\right)^2 > 0 \tag{22}$$

Numerical Example

The reduced deterioration rate, $m(\xi)$, is a function of preservation technology cost ξ such that $m(\xi) = m_o e^{-\delta\xi}$, where δ is the simulation coefficient representing the percentage of increase in $m(\xi)$ per increase in ξ, which means $m(\xi)$ is an increasing function bounded above by m_o.

Crisp Model

Consider $\alpha = 200$, $\beta = 0.05$, $h = 0.5$ ▢/unit, $c = 4$▢/unit, $l = 8$▢/unit, $T = 50$ days, $\delta = 0.1$, $k = 0.07$, $s = 6$ ▢/unit, $c_0 = 100$, $m_o = 0.02$ in appropriate units. The values of different parameters considered here are realistic, though these are not taken from any case study. By using Mathematica 11.1 software, we get the optimum value of critical point $t_1 = 39.1118$ days, preservation technology cost $\xi = ▢\ 41.2491$ and economic order quantity $Q = 24473.9$ with optimum average total cost $TAC(\xi, t_1) = 704.032$. However, the concavities of the average total cost function with respect to t_1 and α are shown in the fig. 2 below.

Fuzzy Model

Suppose $\tilde{\alpha} = (180, 200, 220), \tilde{\beta} = (0.04, 0.05, 0.06), \tilde{h} = (0.45, 0.5, 0.55), \tilde{c} = (3,4,5), \tilde{s} = (4,6,8)$ and $\tilde{l} = (5, 8, 10)$ which are considered as triangular fuzzy numbers and all other parameters are kept same as in crisp model. Then the fuzzy total average cost obtained by (i) Signed distance method is $\widetilde{TAC_s}(\xi, t_1) = 627.09$ with the optimum value of critical point $t_1 = 38.6923$ days, preservation technology cost $\xi = ▢\ 40.4994$ and economic order quantity $Q = 34681.6$, (ii) Centroid method is $\widetilde{TAC_c}(\xi, t_1) = 601.442$ with the optimum value of critical point $t_1 = 38.5524$ days, preservation technology cost $\xi = ▢\ 40.2496$ and economic order quantity $Q = 38084.1$

Sensitivity Analysis

It is very important in an inventory system for a retailer to know the behavior of the system parameters which impacted upon the average total cost function. Retailer should know when one will get minimum expenditure after increase or decrease of the related parameters. Henceforth to illustrate the applicability of the model and to locate some significant managerial ramifications in the vegetable farm houses, we study sensitivity analysis with the variation of different parameters.

Table 1: Sensitivity analysis for parameter $\tilde{h}$

Managerial Insights

The following managerial phenomena are observed from table 1 to table 6:

(i)   From the numerical example, we see that fuzzy model gives best optimal solution as compared to crisp model.

(ii)   From table. 1, we observe that when holding cost $\tilde{h}$ of the vegetable increases keeping all other parameters unaltered, both the preservation technology cost ξ as well as critical point t_1 decreases with the increase of the minimum expenditure $\widehat{TAC}(\xi, t_1)$. In this case Centroid method gives best result as compared to signed distance methods.

(iii)   With increase in deterioration cost parameter $\tilde{c}$ in table. 2, we see that the value of the preservation technology cost ξ as well as critical point t_1 increases with decrease in the minimum expenditure $\widehat{TAC}(\xi, t_1)$.

(iv)   With increase demand parameter $\tilde{\alpha}$ in table. 3, we see that the value of the preservation technology cost ξ as well asa critical point t_1 increases with increase in the minimum expenditure $\widehat{TAC}(\xi, t_1)$.

(v)   With increase in demand parameter $\tilde{\beta}$ in table. 4, we see that the value of the preservation technology cost ξ increases slowly and critical point t_1 increases with decrease in the minimum expenditure $\widehat{TAC}(\xi, t_1)$.

(vi)   From table. 5, we observe that when lost sale cost $\tilde{l}$ of the vegetable increases keeping all other parameters unchanged, the value of the preservation technology cost ξ as well as critical point t_1 increases with increase in the minimum expenditure $\widehat{TAC}(\xi, t_1)$.

(vii)   From table. 6, we observe that when shortage cost $\tilde{s}$ of the vegetable increases keeping all other parameters remains unchanged, the value of the preservation technology cost ξ increases as critical point t_1 increases with the increases of the minimum expenditure $\widehat{TAC}(\xi, t_1)$. However in this case the increase is very slow.

CONCLUSION AND SCOPE

In this paper, we study a kind of deteriorating seasonal products whose deterioration rate can be controlled by investing on the preservation efforts. The public demand always plays a vital role in case of purchasing vegetables, fruits etc. Keeping this in mind we consider that the demand depends upon the stock level. Due to uncertainty nature during the pandemic, the price of the product fluctuates depending upon different related parameters. On focusing the above assumptions we developed this model and found that it gives always profit in the sense of economic as well as humanity. In this conclusion we observe that fuzzy model give minimum expenditure as compared to classical crisp model. Moreover this model is highly essential at the time of lockdown and shutdown of the market during the pandemic COVID-19 in the countries like India and Pakistan. For future research, the proposed model can be extended in several ways. For example, one can extend by adding purchasing cost and selling price. Also one can extend this article by adding some other related parameters like credit policy, inflation etc.

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Table 1: Sensitivity analysis for parameter $\tilde{h}$

$\tilde{h}$	t_1		ξ		$\overline{TAC}(\xi, t_1)$	
	SDM	CM	SDM	CM	SDM	CM
(0.45,0.46,0.47)	45.2274	45.2387	43.292	43.2941	560.993	560.728
(0.46,0.47,0.48)	43.5344	43.5444	42.7496	42.7515	600.614	600.377
(0.47,0.48,0.49)	41.9581	41.956	42.2302	42.232	637.384	637.172
(0.48,0.49,0.5)	40.4903	40.5014	41.7321	41.7337	671.599	671.408

Table 2: Sensitivity analysis for parameter $\tilde{c}$

$\tilde{c}$	t_1		ξ		$\widehat{TAC}(\xi, t_1)$	
	SDM	CM	SDM	CM	SDM	CM
(2,2.5,3)	27.2479	27.2831	33.9814	33.9836	1052.19	1050.93
(2.5,3,3.5)	30.6976	30.7411	35.7703	35.579	954.197	952.643
(3,3.5,4)	34.6766	34.7309	38.7789	38.7874	837.268	835.327

Table 3: Sensitivity analysis for parameter $\tilde{\alpha}$

$\tilde{\alpha}$	t_1		ξ		$\widehat{TAC}(\xi, t_1)$	
	SDM	CM	SDM	CM	SDM	CM
(200,220,240)	39.1489	39.1483	42.2002	42.1930	770.041	770.034
(220,240,260)	39.1815	39.1810	43.0901	43.0841	835.990	835.984
(240,260,280)	39.2089	39.2086	43.9070	43.9019	901.868	901.863
(260,280,190)	39.2300	39.2289	44.5760	44.5429	959.464	956.720

Table 4: Sensitivity analysis for parameter $\tilde{\beta}$

$\tilde{\beta}$	t_1		ξ		$\widehat{TAC}(\xi, t_1)$	
	SDM	CM	SDM	CM	SDM	CM
(0.040,0.042,0.044)	35.3123	35.3157	40.9752	40.9781	793.841	793.761
(0.042,0.044,0.046)	36.1965	36.2007	40.9971	40.9998	772.935	772.850
(0.044,0.046,0.048)	37.1254	37.1284	41.052	41.6546	751.002	750.909

Table 5: Sensitivity analysis for parameter $\tilde{l}$

$\tilde{l}$	t_1		ξ		$\widehat{TAC}(\xi, t_1)$	
	SDM	CM	SDM	CM	SDM	CM
(5,6,7)	32.8314	32.8306	37.7498	37.7347	596.759	595.783
(6,7,8)	35.9743	35.9738	39.5598	39.5472	654.778	653.805
(7,8,9)	39.1105	39.1101	41.2173	41.2067	701.118	700.147
(8,9,10)	42.2415	42.2411	42.7464	42.7373	735.802	734.832
(9,10,11)	45.3682	45.3679	44.1656	44.1578	758.847	757.878

Table 6: Sensitivity analysis for parameter $\tilde{s}$

$\tilde{s}$	t_1		ξ		$\widehat{TAC}(\xi, t_1)$	
	SDM	CM	SDM	CM	SDM	CM
(4,5,6)	38.876	38.876	41.1291	41.1291	700.934	700.928
(5,6,7)	39.118	39.1118	41.3681	41.2489	711.931	714.564
(6,7,8)	39.3476	39.3476	41.3679	41.3678	735.802	734.832
(7,8,9)	39.5834	39.5834	41.486125	41.48606	758.847	757.878
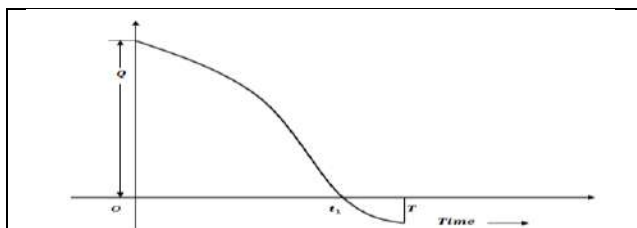
Fig. 1 The graphical representation of the inventory system: inventory versus time

Fig.2 Crisp Model

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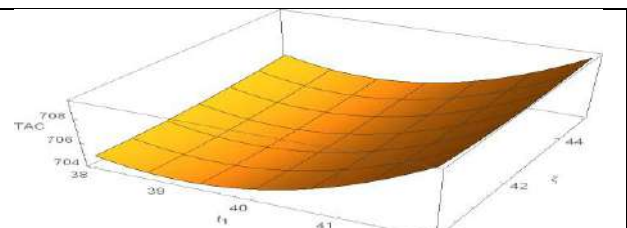
Isolation and Identification of Amylase Enzyme Producing Thermophilic Bacteria from Lasundra Hot Spring

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ABSTRACT

Amylases are one of the most important enzymes in biotechnology right now. The Thermostability of amylase generated by thermophilic bacteria increases the starch degrading industrial process in the starch industry. The goal of this research was to isolate and identify a thermophilic amylase-producing bacterial strain from a hot spring near Lasundra, Gujarat, India. The amylase activity test was performed on all isolates after the thermophilic bacteria were isolated at 59 ̊C. The strain KG 1 has the highest amylase activity among the few isolated amylolytic strains on starch agar media. Based on 16s rDNA sequence homology, phylogenetic analysis suggests that the isolated strain belongs to the genus *Bacillus* and is designated *Bacillus cereus.*

Keywords: Thermophilic, amylase, hot spring

INTRODUCTION

Thermophiles are microorganisms that have evolved to thrive in hot environments. They can resist such intense heat because their structural components and biomolecules, including proteins, lipids, enzymes, ribosomes, ribonucleic acid, and deoxyribonucleic acid have undergone various alterations (1) Enzymes must be stable under process conditions for manufacturing use. Chemical catalysts are generally favoured over enzymes. According to the result,

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thermophilic bacteria are thought to be a good alternative source of thermostable enzymes. (2) The amylase family of enzymes (a-amylase, ß-amylase, and gamma amylase) has a wide collection of commercial applications, including food, fermentation, textiles, paper, and detergents. When in fact they can come from flora, fauna, and microbes, fungal and bacterial amylases are primarily employed in mass production in the industry due to their greater production rate, thermostability, reduced time and space requirements, and ease of process modification and optimization. (3) Thermophilic bacteria are microbes that live and thrive in temperatures ranging from 45 to 122 ○ C. (4) Numerous enzymes are utilized in bioindustries; for example, protease and amylases are used together in many sectors, including food, detergents, and pharmaceuticals. (5).

As a result, these thermophilic microbial sources meet the needs of the detergent, pharmaceutical, culinary, textile, and R&D industries. Thermostable catalysts are steady and operative in high-temperature environments, opening up novel biocatalysis and biotransformation possibilities. The principal sources of thermophiles are geothermally heated places on Earth, such as hydrothermal vents, hot springs, geysers, and compost. There are more than 350 springs in India, as stated by the Indian Geological Survey, which is categorized based on their geo-tectonic configuration. (6) Moderate thermophiles (growth optimum: 50–60℃), extreme thermophiles (growth optimum: 60–80℃), and hyperthermophiles (growth optimum: 80–110℃) are the three types of thermophiles. Thermophiles have been secluded from many ecological zones on the planet (for example; geysers and the deep sea). Members of the Archaea genera *Pyrobaculum*, *Pyrodictium*, *Pyrococcus*, and *Melanopyrus* have the highest growth temperatures (103–110℃); within Fungi, the Ascomycetes and Zygomycetes classes have high growth temperatures, and in bacteria, *Thermotoga maritime* and *Aquifex hygrophilous* have the maximum growth temperatures (90 and 95℃ respectively). Gram-positive and Gram-negative thermophilic microorganisms exist in both aerobic and anaerobic environmentsand some of them can generate spores (7).

After lignocelluloses and chitins, starch is the broadest polysaccharide generated by plants as energy storage. Amylases are extracellular enzymes that catalyse the hydrolysis of internal 1,4-glycosidic bonds in starch to dextrin and other small glucose-containing carbohydrate molecules. They are categorized into three categories derived from the cleavage site: alpha-amylase, beta-amylase, and gamma-amylase. The alpha-amylase acts at random locations near the starch chain, beta-amylase hydrolyses the second alpha-1,4-glycosidic bond, cut off two glucose units, whereas gamma-amylase cleaves alpha-1,6-glycosidic linkages, as well to cleaving the last alpha-1,4-glycosidic linkages at the non-reducing end of amylose and amylopectin, having the result that glucose production. Amylases have a broad range of implementation in the starch processing, detergent, alcohol, textile, food, paper, and pharmaceutical industries which has drawn the attention of the worldwide enzyme industry. The usage of amylase enzyme in industry especially the pharmaceutical industry, necessitates full purification. Plants, animals, and microbes can all provide amylases. Microorganisms are the most common source of amylase production because of their optimal growth needs, accessibility, efficiency, environmental friendliness, and cost-effectiveness when compared to other resources including animal and plant sources (8).

The usage of amylase is growing in popularity around the world. Despite this, there is a lot of interest in finding enzymes with better featureslike thermostable raw starch degrading amylase for industrial use and cost-effective manufacturing processes. Contamination and product batch-to-batch variability is a serious difficulty in an industry that operates in moderate settings, necessitating us to perform the fermentation activity at severe temperatures where contamination is almost non-existent (9). The main expected outcomes of this study will suggest that hot springs include a diversified range of thermophilic bacteria that can serve as possible reservoirs for the development of thermostable amylase, which has a wide range of applications. The current research focuses on (i) isolating thermophilic bacteria from hot springs and (ii) screening the isolates for the production of industrially significant amylase.

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MATERIALS AND METHODS

Isolation Site and Cultivation Conditions

Water samples were collected at the depth of 10 to 12 feet in a sterile thermos bottle from Lasundra hot water springs and brought to the research laboratory in aseptic conditions. The maximum temperature of the central reservoirs was 50 ͦ C the nearby reservoirs have a temperature varying between 42 to 47 ͦ C. Bacteria were isolated by serial dilution and spread plate method. A volume of 0.1 ml of dilution was transferred to Nutrient agar (0.5% peptone, 0.1% yeast extract, 0.5% sodium chloride, 1.5% agar, pH = 7.4) The sample were spread uniformly using a glass rod, and incubated at 59 ͦ C for 24 hours. The bacterial isolates were further subcultured on nutrient agar order to pure culture. Pure isolates were refrigerated at 4 ͦ C for further studies.

Screening of Amylase Producing Bacteria

Microbial isolate streaked starch agar plates, which were then incubated at 59 ͦ C for 24 hours. After incubation, the starch agar plates were flooded with a 1%iodine solution (freshly made). A clear zone of hydrolysis surrounding the growth sites indicated a positive result, whereas the presence of blue colour around the growth suggested a negative result.

Identification of Amylase Producing Bacteria

Under the microscope, the colony morphology of the isolates was examined in terms of colour, shape, size, colony nature, and pigmentation.

Molecular Characterization

On the basis of 16s rDNA sequence analysis, the molecular identification was carried out. A pure culture of KG 1 was used to isolate genomic DNA. The 16SrRNA-F and 16SrRNA-R primers were used to amplify a 16S rRNA gene fragment. When resolved on an agarose gel, a single distinct 1500bp PCR amplicon bond was found. Barcode Biosciences, based in Bangalore, India, carried out the sequencing. The 16S rRNA gene sequence was BLAST against the NCBI GenBank database's "nr" database. The first ten sequences were chosen and aligned with Clustal W, a multiple alignment software programme, based on their maximum identity score. MEGA 10 was used to create a distance matrix and a phylogenetic tree.

RESULT

Isolation of Amylase Producing Bacteria

Based on colony shape, a total of eight bacteria strains were identified. The generation of amylase by starch hydrolysis was the primary criteria for all isolates. The starch agar was hydrolysed by three of the eight isolates tested. After pouring iodine solution over starch agar plates, the isolates KG 1, KG 2, and KG 3 revealed the most clear zone, indicating amylase activity.

Morphological Characteristics

Table 1

Molecular Identification

KG 1 was found to be Bacillus cereus, showed high similarity based on nucleotide homology and phylogenetic analysis. Evolutionary analyses were conducted in MEGA11 .

Evolutionary Analysis By Maximum Likelihood Method

The evolutionary history was inferred by using the Maximum Likelihood method and Tamura-Nei model [10]. The tree with the highest log likelihood (-2150.35) is shown. Initial tree(s) for the heuristic search were obtained

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automatically by applying Neighbor-Join and BioNJ algorithms to a matrix of pairwise distances estimated using the Tamura-Nei model, and then selecting the topology with superior log likelihood value. This analysis involved 11 nucleotide sequences. Codon positions included were lst+2nd+3rd+Noncoding. There were a total of 1461 positions in the final dataset. Evolutionary analyses were conducted in MEGA11 [11]

DISCUSSION

In this study, an isolate of thermophilic bacteria was recovered and identified from a hot spring in Lasundra, Gujarat. The temperature of hot springs is frequently more than 40 degrees Celsius, making geothermal areas that are favourable homes for thermophilic organisms limited to a small number of locales. Living organisms must deal with extremes in temperature, low humidity, and few nutritious components in these environments. To adapt to such stress, some individuals adopt survival tactics. Among the few isolates amylolytic strains on starch agar medium, the strain KG 1, KG 2, KG 3 showed amylase activity and KG 1 showed best amylase activity. The genus bacillus was isolated from explored site, the presence of bacillus in sample location could be due to the ability of this genus to more at high rates and their resistance to harsh environment conditions, in addition to its adaptation for hot surrounding. In the present study, the result obtained from 16s rDNA sequencing for KG1 were analysed by BLAST alignment of these isolate to GenBank sequence, the phylogenetic tree was constructed. In this study is the isolation of strain KG1 , which was identified by 16s rDNA as bacillus cereus. One of the important potential of thermophilic micro-organisms is their enzymatic activities; due to their ability to function under sever conditions, this will intensify their importance in industrial and biotechnological areas.

ACKNOWLEDGEMENT

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Table 1: Morphological Characterisation

CHARACTRISTICS	KG 1	KG 2	KG 3
COLOUR	YELLOW	WHITE	YELLOW
SHAPE	CIRCULAR	CIRCULAR	CIRCULAR
EDGE	ENTIRE	ENTIRE	ENTIRE
TEXTURE	SLIMY, MOIST	OPAQUE	OPAQUE
ELEVATION	FLAT	CONVEX	CONVEX

KG 1   KG 2   KG 3
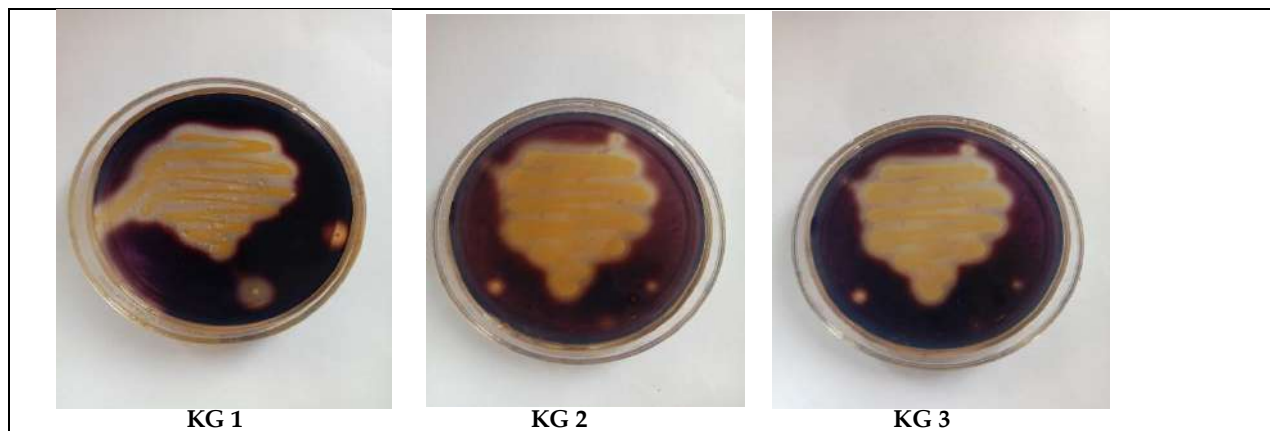
Fig.1 : Amylase activity test

gDNA   16SrRNA amplicon
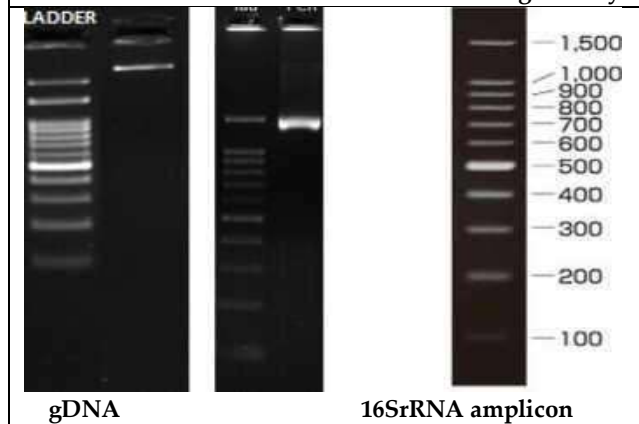
Fig.2 : Ladder Specification   **Fig.3 : Phylogenetic Tree**
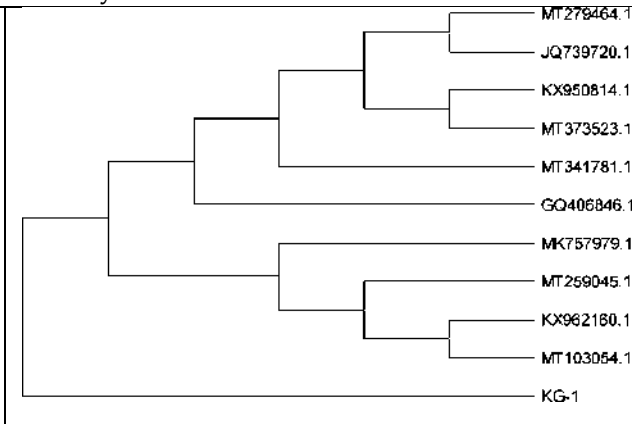
REVIEW ARTICLE

Personalized Medicine: A New Era in Dentistry

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ABSTRACT

As today's world is heading towards the concept of individualization, so is it also pertaining towards one's treatment or medication. Personalized medicine is a breakthrough in the field of science that tailor a treatment as individualized as the disease. It is a newly emerging but rapidly progressing field of health care where a practitioner is able to select a treatment plan according to patients unique genetic, environmental, and clinical profile, that minimize harmful side effects and provide a better result. It can be cost effective when compared with a normal approach to treatment for disease. Dentists and physicians have long recognized variations between and among patients, and have customized care based on each individual's health history, environment, and behaviour. In this article, we present an overview of the opportunities and challenges that influence the oral health in personalized medicine. We highlight selected research advances that are solidifying the foundation of personalized oral health care, elaborate on their impact on dentistry, and explore obstacles toward their adoption into practice. Practice of personalized medicine will surely ensure quality, customized, and effective oral health care for all.

Keywords: Personalised Medicine, Genetics, Dental application, Drug design.

INTRODUCTION

Most of the oral diseases arise due to the complex interaction between genetic, biological, behavioural, environmental and various other factors. Because of differential expression and interface of these factors, patients differ in their susceptibility and expression of disease. There have been many advances in genomics, proteomics, metabolomics, and molecular medicine in the last two decades. Personalized medicine customizes the health care treatment by the usage of these advances along with clinical profile and other relevant data. Personalized medicine is

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also known as stratified medicine as patients are stratified based on information given, and then therapy is classified according to characteristics of each level.(1)The 21st century vision of personalised medicine is to offer 'the precise drug, with the precise dose at the precise time to the precise patient'. Effective application of personalised medicine depends on the accessibility of accurate diagnostic tools which allow for the finest selection of therapeutic product to improve the patient outcomes. The uprising of personalised medicine has created a rewarding opportunity for pharmaceutical companies to develop molecular-targeted medications, but in an optimised use and repurposing of existing medication and combination therapies. Finally personalised medicine will modify the approach to diagnosis and treatment, and this will lead to increased contribution of the individuals during and after treatment.(2) A patient who walks in for a routine check should be administered treatment tailored to his need.

BENEFITS
Personalised medicine has many benefits including the potential to offer better medication selection and targeted therapy, decrease any adverse effects, improve patient compliance, change the goal of medicine from reaction to prevention, develop cost effectiveness, and increase patient confidence post-marketing by approving new therapeutic strategies and altering the perception of medicine in the treatment. Other important benefits include: better medication selection, safer dosing options, and improvements in drug development

APPLICATION IN DENTISTRY
Most of the dental/oral diseases: dental caries, periodontal diseases, oral cancer, chronic orofacial pain, malocclusion, cleft palate, occurs as result of complex interaction of genetic, biological, lifestyle and environmental factors. The link between oral and systemic diseases is a well-known fact. (3) and perio-systemic linkage is very common (Fig1)

Periodontal applications and interleukin-1:
Smoking, diabetes, and certain genetic variations are the major risk factors associated with severe periodontal disease. As per our current oral care practice, all adults are assumed to be at equal risk, and given similar treatment type of preventive and interceptive care. Thus, the opportunities to prevent disease in high-risk patients and needless services given in low-risk individuals, health care costs in many countries are unsound. As per the recent literature, periodontitis is considered as a polymicrobial infection, with elevated levels of proinflammatory cytokines: interleukin-1 (IL-1),playing a major role in the pathogenesis of disease(4). William Giannobile et al (5) utilized personalized medicine approach in his study, in which he evaluated tooth loss in 5117 periodontitis patients, over a period of 16 years. Patients were classified based on three risk factors - smoking, diabetes and interleukin-1 (IL-1) positive genotype. They concluded that, in low-risk patients, tooth loss had no impact on whether the patient visited the dentist once or twice annually but in high-risk patients, two preventive visits per year significantly decreased the number of patients who had tooth loss problems. Hence, it was concluded that professionals should customize their approach to preventive care based on initial risk assessment and by using personalized preventive care and treatment can be given for the individuals who are at higher risk for disease.

Interleukin-1:
The presence of IL-1 positive gene is associated with increased inflammatory response. Patients who are genetically positive for IL-1 gene show higher prevalence and increased severity of periodontal disease and can be used as a marker for those patients who have any other risk factors for periodontal disease. Dentists should thus consider doing this test as a part of a comprehensive periodontal treatment, available under the trade name of Peri Predict. At present, this is the only genetic susceptibility test used for periodontal diseases. (1,6)

Management of dental caries
Dental caries is the most common oral disease and being multifactorial, many factors including environmental, behavioural, dietary habits, micro flora, host factors: oral hygiene, salivary composition, morphology of the tooth and genetic factors are associated with the incidence of dental caries. preventive measures include use of small molecules, aimed at blocking or weakening enzymes that causes the bacteria to form a bio film or attach to the tooth surface. The different risk factors associated with dental caries varies between individuals and therefore can be used

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to categorize the patients and individualise the therapy.(7) A caries risk assessment should also focus on factors that may cause the patient's ability to maintain oral hygiene, morphological aspects of tooth like deep fissures & pits, crowding, restorative margins and use of oral appliances.

Orthodontics

Skeletal and dental malocclusion has a genetic background, but always stood out as topic of debate. The application of genetics to clinical orthodontics is a major challenge to develop a comprehensive and detailed phenotypic classification. Personalized orthodontics is tailed to an individual in whom the genomic and clinical data can be utilized to anticipate the development of malocclusion(s) and consequently, treatment plan can be implemented(8).Mandibular prognathism (Class III) and cleft lip & palate are usually genetic in origin. However, the genetic background alone does not reduce the challenge in diagnosis and treatment planning. Further aspects like the type of craniofacial growth can also help in the planning of both the type and timing for orthodontic and surgical treatment.

Cancer management

Detection of a disease-causing mutation among a family can tell the most risk individuals whether they are at higher risk for cancer and can begin individualized prophylactic treatment. The cancer management focused on individualized treatment based on tumour stage, subtype, and histology. Recent developments in the omics tools: genomics, epigenomics, transcriptomics, proteomics and metabolomics are novel paths in the course of biomarker detection for rapid and early diagnosis of oral cancer(9). Data obtained from genetic studies gives a far more refined tumour classification based on signalling pathways that can be targeted more precisely. Molecular techniques are also helping to predict the lesions which are likely to undergo malignant transformation through a better perceptive of wide range of proteins and transcription factors that help in the epithelial mesenchymal transition. Thus, customized treatment helps to better define the prognosis in these patients and for better treatment options that are most likely to succeed. Identification of tumour markers helps in identification in the chance of recurrence. Several researches based on protein expression and nucleic acid analysis show more accurate prediction of patients at risk for any recurrence of oral cancer.

Molecularly targeted therapies are in the process and are being tested for the treatment of head and neck squamous cell carcinoma. Among the targeted therapies the drugs under study is tyrosine kinase inhibitor such as imatinib, developed to treat chronic myeloid leukaemia specifically inhibits the Ableson tyrosine kinase protein, which is strongly correlated with disease. This is an example of "rational drug design" developed based on knowledge of disease pathophysiology. Other examples include epidermal growth factor receptor (EGFR)-directed drugs, such as monoclonal blocking antibodies (cetuximab) and EGFR-tyrosine kinase inhibitors (gefitinib, erlotinib, lapatinib), which have been tested in clinical trials(10). Multiple contributors influence the personalized phenome, including metabolomic and proteomic components that are modified by extrinsic and intrinsic environmental factors, under the genomic and epigenetic control. These factors have to be taken into consideration when looking into personalised medicine (11).

Management of acute and chronic or facial pain:

Identification and monitoring of individual genetic variation may enable dentists to modify pre-operative and post-operative pain management such that it is more effective. Genetic variation also plays an important role in an individual's insight of pain, thus will enable the dentists to know about the receptiveness to chronic or facial pain or temporo-mandibular joint disorders (TMD). Researches ongoing in TMD disorders, includes genetic, psychosocial, and pain amplification data, that the clinicians could incorporate into health-history related questionnaires, allowing for diagnosis of patients who shows the most abnormal pain sensitivity and psychosocial profile. (10)

Oral infectious diseases

A personalized approach for treating oral infectious diseases is progressing quickly, based on the research activities. Such data provides brand new methods for disease prevention. Advances in imaging shall provide better

understanding of oral bio-film development, organization, and composition. This will provide the design of biofilm-manipulating treatments (10).

CHALLENGES AND FUTURE SCOPE

In many ways, personalized medicine is indeed here, but many improvements are needed to authenticate its routine and efficient use in oral health care. Current scientific and technological gaps include: classic linkages between both biomarkers and genotypes and clinical results, cost-effective and non-invasive imaging technologies for the detection of pathologies of the oral-craniofacial complex; illumination of the association between and among the environment, microbiome, and genetics, more precise prediction between disease risk and drug response , improved drug design and delivery , an appropriate use and access to electronic health records. Many genetic variations exist and in addition how one may respond to a drug is not only determined by only one gene but also by many genes interacting with each other (10).

On the educational aspect, future health workers will need to be well informed in the scientific underpinnings of personalized care. As genetic testing becomes more common, it is unclear how health care professionals including dentists, will be able to understand them. Dental institutions will need to incorporate genetics and genomics into their normal curricula, and dentists will need to be updated with the changing technologies. But the main element of the success of personalized medicine is the public awareness and acceptance of the benefits and risks of personal genome sequencing. Socio-ethical and legal barriers will prevail in the field of genomic medicine, stem cell therapy, and other molecular approaches to health care, and measures are needed to be addressed considerately with public participation. This reality underscores the need for a deeper understanding of the public's unique educational requirements, as well as for genomic education efforts including various communication channels and approaches such as social media and comprehensive educational campaigns.(10)

CONCLUSION

Personalized medicine has great uses in diseases like periodontitis, rampant caries, cancers, where the treatment options are uncertain. Thus, usage of personalized medicine to routine dentistry is closer than one might think.(1)It also provides opportunity to develop the value of currently approved drugs with limited market. Personalised medicine has the prospective to fulfil the prerequisite to improve health care by sinking the healthcare costs, drug-improvement costs and time. This revolution can only be achieved by equal contribution by the patient and consumers participating in clinical trials, investors and innovators to develop smart tools and analyse the genetic information and support essential revolutions in policy and regulation, practitioners to understand the disease at the molecular level, academic researchers for innovative research to develop new insights at the molecular basis of disease and to support target-based drug development and secure patient information, and entrepreneurs by developing new business models, new diagnostics tools, target therapy and other customised treatment protocols. Personalised medicine has the potential to have a positive impact on the healthcare system. In future, with use of the personalized approach, each individual will receive their full genomic information from their day of birth into an individual medical record. This information would allow clinicians to apply more effective healthcare treatment based on patient exposure to any different diseases. (2)

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Fig.1. Application of Personalized Medicine in Dentistry

RESEARCH ARTICLE

Sentiment Polarity through Sarcasm Identification and Detection in Tweets

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ABSTRACT

Sentiment Analysis and Opinion Mining is the gaining augment allurement due to enlarged unsolved challenges. The evolution of web 2.0 also re-percussed the growth of different social networking sites, profiting the users, to easy access of data which can be analyzed to obtain the functional information. Identification of sarcasm is a colossal task in sentiment analysis. Detecting the sarcasm is an indispensable commission to obtain the real polarity of the text. The research work focuses on ascertaining of tweets for sentiment polarity through sarcasm detection via machine learning classifiers such as Gaussian Naive-Bayes, Logistic Regression(LR), Linear Support Vector Classifier(LSVC), Random Forest Classifiers. The tweets are collected through twitter API and pre-processed and further structured and analysed. Linear SVC and LR has produced better results in terms of accuracy compared to other classifiers .

Keywords: Sarcasm, Text mining, Sentiment Analysis, Machine Learning

INTRODUCTION

In the times of yore communication was an important and laborious task; long voyages were taken place just to transmit or gain pittance of information, as evolution in software, hardware and internet technologies gave emergence to anytime anywhere access to information influenced the communication pattern. People could communicate with an easy access, this lead into another advancement of social networking sites, where people could

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share their opinions, interests, activities, current issues and so on. Peoples' opinions on certain facts have become a key factor for making decisions in many circumstances such as deciding on policies, making business decisions, e-governance etc. There were surveys, experiments and interviews were conducted just to find out about the opinions of the people before concluding on various subject matters, but with the help of social networking sites opinions of individual or group of people are available under one roof, which can be analyzed in order to generate useful information, which is called as knowledge in data mining[1]. With the ease of development of smart phones and online platforms to share a common interest and opinion of oneself the social networking services become an expedient tool, among most common ones' are Facebook, Twitter, LinkedIn, Instagram, Tumblr etc. The social networking users are expected to be 4.41 billion by 2025 according to the statista. Twitter is micro blogging site yielding to convenient expression of opinions and sentiments of users through short message services[3]. Now a day's twitter data is sought to identify the public opinion on political, commercial, national, international, entertainment, celebutantes and business views. The objective of the research work is to extract the tweets from twitter API and remove the noise by ascertaining the tweets for sentiment polarity via sarcasm detection via different machine learning classifiers and yield the results in measures of accuracy. The research paper is structured as following sections. Introduction followed by motivation, related work which describes the some of the relevant works done recently, and the next section describes about the methodologies used in detail along with the results discussing different performance matrices followed by conclusion.

Motivation

The challenges in sentiment classification is sarcasm. Sarcasm is stating or communicating just the contrary to what one exactly have in view. Spontaneous detection of sarcasm is still infant in the research field of sentiment analysis and natural language processing. The traditional methods such as n grams, emoticons identification, pragmatic engineering were not enough to detect the sarcasm, various new techniques are explored. Sarcasm is way of defining negating emotions positively. For example: "I am feeling super motivated" instead of saying "I am demotivated", another example is "I love pain", but actually the same person wants to specify that he is going through hard times. Sarcasm texts are challenging to Identify as the spoken words can be easy to identify compared to written sarcasm text. Sometimes it is very essential to understand the context and background of the statement, also sarcasm specific statement will be limited to a specific set of users, if the patterns are closely watched in contextual phenomenon. The motivation behind the work is unsolved challenges such as strenuous in identification of irony in a text or sentences, yet to receive accurate methodologies of in NLP. Many researchers are focusing on satirical identification for better sentiment classification are yet to build a model with accurate results in the field of computer science.

Related Work

The inclination of expressing opinions in micro blogging sites are expanding stupendously, Sentiment analysis are illustrated via mapping of tweet location in two approaches by Ingle et al. [4], one is Tidy verse package in R tool and the second is deep learning. The tweets are based on demonetization and abolition of article 370, the stages are tokenization and categorization. The tweets are tokenized and each token is unique, which are tagged as positive, negative and neutral. For each of these sentiments, the score is computed, In the second approach, deep learning algorithm is used, the text is pre-processed and weighted through TF-IDF model. Text to vector conversion is performed by Glmnet package. The tweet location is plotted with GIS methods. Deep learning model was more promising than the Tidyverse and yielded 87% accuracy. The future scope of the work is exploring the same for vernacular languages along with different social media platforms with more enlarged data sets. The research work does not disclose the details about number of tweets and so on, whilst Ashima et al. [2] explored the SVM for sarcasm detection on twitter data, the work consists of find the polarity in tweets using unigram and interjection features through Support vector machine with polynomial kernels. The unigram feature extraction process involved in fragmenting the tweets into token such as fabricating the tweets to single and unique words, and, an interjection words had a direct relation with person's feeling, further it was grouped to diversification of emotions. According to their results, the accuracy was 91% using proposed model, the cross validation method was used to affirm the results. Shubhadeep Mukherjee [10] has contributed on sarcasm detection in tweets through fuzzy clustering

methods and naive bayes, using hashtags tweets are classified as sarcasm and not sarcasm sentences, through naive bayes classifier, ironic tweets are differentiated from non-ironic tweets. The research work had contribution in a new approach in sarcasm identification and detection, the feature selection part involves various features such as part of speech, pos n-grams along with author of the tweet, both supervised and unsupervised methods illustrate the different results by concluding features that are independent of text, increased the accuracy in prediction, along with authorial feature pattern perform finer than part of tags and part of speech n-grams, together with Naive Bayes classifier as number of tweets are too less for the computation. Jose Antonio [5] had worked on new upheaval of fast opinion mining via information retrieval techniques, the research work focuses on automatic learning of model for sentiment analysis, although this kind of work has evolved from past researchers, finding the polarity from the information retrieval process with a help of document analysis and language independent sentiment analysis, appear to be credulous for the sentiment analysis. The two methods IRSAC rank and IRSAC agr had been built for extraction of features, IRSACrank utilizes the raw data, IRSAC rank encompasses all the data required by IRSACarg, and both models complement each other.

Bala Durga et al. [6] proposed a new methodology for Sarcasm Detection in Twitter by classifying the data into sarcastic and non-sarcastic classes, the Adaboost algorithm was selected to identify the weak and strong statements recursively, the initial process was to obtain data and pre-process it, then select the features and classify through Naive Bayes and finally categorize them as sarcastic and non-sarcastic tweets through proposed model. New alteration in machine learning algorithms also yielded to finer techniques and findings in natural language processing, sarcasm is a challenging task for humans as it requires good wit to understand the statement and analysis the sarcasm statement, Le hoang et al. [7] had proposed a model for Sarcasm Detection via model called Soft attention-based bidirectional LSTM Model with Convolution network, fifteen thousand sarcastic and twenty five thousand non-sarcastic messages were detected through sarcasm detection tool, an exploratory investigation for training and test data performed via LSTM, convnet, and bidirectional LSTM without or with attention was observed and the results were computed as convolution model outperforms sarcasm classification as 97% for twitter dataset. Aniruddha Ghosh et al. [9] tossed Timely Sarcasm Detection for Contextual and Personal, giving enough evidence of same person tweeting the sarcasm tweets over and again based on the psychological context of the user. Ravinder Ahuja et al. [8] had proposed a study of comparative different algorithms for Sarcasm Detection on behavioral approach via applying twelve different classification algorithms such as gradient boosting, Guassian Naive Bayes, Adaboost, Decision Tree, Extra Trees, Random Forest, SVM (rbf- kernel), L1 regularized Logistic Regression, Gradient Boosting, L2 regularized Logistic Regression, Bagging Model, K-neighbors hybrid and K neighbors also four types of datasets to obtain the accuracy of the algorithms were selected.

METHODOLOGY

Social Networking has succored to Cumulative decision making, which plays an important role in classic decisions for industries who look upon the diversified users. Hence, the proper classification and approach is essential to identify opinions of others especially when the sentences are sarcastic and ironic. Detecting sarcastic or irony emotions in the given text is exigent [11]. The proposed model is implemented in R programming and Python. In R studio included the packages like twitteR, sentiment, e1071, caTools, RTextTools, caret, Sentimentr and in python pandas, numpy, nltl.stem.porter, keras were some of the interfaces for analysis.The Figure 1 shows the flow of work carried out. Data is obtained from Twitter microblogging through twitter API, data is pre-processed, feature selection is done via IF-IDF, model is trained and testing is carried out by dint of ML algorithms and results are tabulated.

Data: The data was collected from twitter API in R tool, through the keywords Bengaluru Traffic in English Language, every time the number of tweets size were entered as 2500, and based on the availability of the tweets the API would return some tweets, altogether 20500+ were collected over a period of time and pre-processed, and further classifications were done. The pre-processing involved the of removing unwanted data from the downloaded columns, removing of slangs, misspelled words, hashtags, user mentions, control and special characters,

whitespaces, punctuations and retweets were carried out in the data pre-processing stage. The figure 2 describes the sample sarcasm tweet from twitter.

TF-IDF

After this process, vectorization is carried out, in which, Term frequency- Inverse Document Frequency technique is applied, the text is converted into significant number, where it quantifies, how many elements are instance in the given document present.

The formula for TF-IDF is represented as,

TF-IDF = TF(t,d) x IDF (t)

TF describes the term frequency, i. e., the aggregate of occurrence t seen in the document d

IDF represents the inverse document frequency, count of document accommodating the term t.

$$IDF = log \frac{1+n}{1+df(d,t)} + 1$$

$$TF\text{-}IDF = TF(t,d) \times log(N/df_i)$$

The process of TF-IDF is represented in the figure 3, which simplifies the process of finding the TF-IDF from calculating TF, IDF, on the stemmed data.

Bayesian Sentiment classifier

Bayes classifier uses Bayes theorem to classify the given features and predict the output. Bayesian classifiers has produced better outcome in terms of accuracy and speed when the dataset is huge. The classifier assumes the independence of the attributes with respect to the given class. The below equation (1) illustrates the Bayes theorem.

$$P(A/B)=(P(B/A)*P(A))/P(B) \tag{1}$$

According to equation 1, P(A/B) is posterior probability, P(B/A) is likelihood of evidence, P(A) is prior probability, P(B) id probability of evidence. In Bayesian Sentiment classifier, sequence of text is read as document and reconstructed into raster of features with values as $(f_1,f_2,\ldots\ldots f_n)$. The sentiment polarity Si, can be obtained from feature values, which is represented as

$P(S_i \mid f_1,f_2,\ldots.,f_n)$

$\log(P(S_i) + \log(P(f_i \mid S_i))$

Si is used to determine the sentiment polarity.

Bayesian Classification

Step1: Assumption of independent attributes

$\{f_1,f_2,\ldots\ldots f_n \}$

$P(S_i \mid f_1,f_2,\ldots.,f_n)$

Step2: Assumption of dependent variable: Si

Step 3: Finding the likelihood of the probability

$P(Si \mid f_1,f_2,\ldots f_n)=P(f_1 \mid Si)P(f_2 \mid Si)P(f_3 \mid Si)*P(Si)$

$$P(S_i|f_1,f_2,\ldots f_n) = \frac{P(f_1 \mid S_i)\, P(f_2|S_i)\ldots.P(f_n \mid S_i) * P(S_i)}{P(f_1)P(f_2)\ldots..P(f_n)}$$

$P(S_i) \prod_{i=1}^{n} P(fi| S_i) / P(f_1) P(f_2) \dots P(f_n)$

$S_i = \text{argmax}_y P(S_i) \prod_{i=1}^{n} (f_i | S_i)$

The argmax gives the argument with the highest value from the function.

The regular expressions (RE) was applied to pre-process the data, after the data is pre-processed, features are extracted from the data. Followed by stemming of features, stemming is the process of exterminating the prefixes and suffices in the text unit. In other words, only the root of the word will be considered for data processing. For example: the word "waiting" will be stemmed as wait.

Linear Support Vector

Support vector machines are used both for classification and regression. Linear Support Vector Classifier is used to fit the data on a hyper plane, the kernel is the main component for classification. Kernel converts low dimensional to high dimensional to separate the nonlinear data. LSVC model is well suited for text classification. The main objective of SVC is to find the line which augment the minimum distance to the line. Support vectors and margins are playing important role in terms of finding the hyper plane.

Hyper plane can be represented as

$w.f + b = 0$

If W is a weight vector, f is features, b is bias, then,

$w_0 + w_1 f_1 + w_2 f_2 = 0$

Hyper plane which is defining the sides are

H1: $w_0 + w_1 f_1 + w_2 f_2 \geq 1$ for $y_i = +1$

H2: $w_0 + w_1 f_1 + w_2 f_2 \leq 1$ for $y_i = -1$

Features which fall on H1 and H2 are support vectors.

K is Kernel, 'x' is the feature vector, $\emptyset$ is non linear mapping. Linear kernel is used for classification. It finds the gap between two classes.

$K(x,x) = \emptyset(x) T \emptyset(z)$

Random Forest Classifier

The number of decision tree classifiers are used to estimate the sub samples of the data in random forest classifier, it is an ensemble based learning model. The random forest tree is made of decision trees, features are built based on the hierarchical structure of the trees. More the number of trees, more the accuracy.

The number of decision trees that operate as ensemble, every individual class predict the model, class with highest predicted becomes model's prediction. In a decision tree each attribute will be considered as node branch as end result. Leaf denotes a class prediction as illustrated in the figure 4.

Random Forest

Step 1: In a random forest, X random records are taken, if y is the dataset size

Step 2: Based on records/samples decision trees are constructed

Step 3: Output is assembled from decision trees.

Step 4: Based on highest ballot the output is considered.

Logistic Regression

Logistic Regression employs a logistic function which is used to find the dependent variable given set of independent variable. The data is classified for discrete datasets for binary or multiclass classification. Logistic regression exerts similar to support vector machine except marginal lines are absent. According to figure 5, if the best fit line divides the two sets of data keeping the distance between the points and straight line is minimum, optimum regression is obtained.

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The slope represents the best fit line. It is represented by

$$y=mx+c \qquad (2)$$

In the equation 2m stands for slope, x is data points and c is intercepts.
The same equation can be written as,

$h\emptyset(x) + \emptyset_0 + \emptyset_1 x$
whereas $\emptyset_0$, $\emptyset_1$ are parameters, $h\emptyset(x)$ are hypothesis.
It contains one or more alloy of independent variables termed as

$y = w^T x + b$
$l = \log_b \frac{p}{1-p} = \beta_0 + \beta_1 x$

if $h\emptyset(x) \geq 0.5$, then the value 1 will be assigned for features, else if it is ≤ 0.5 then 0 is assumed. That is, using logistic function the values are retained between 0 to 1.

Since the curve remain to logistic, the sigmoid function or logistic function is employed for predicting the probabilities.

$$\alpha(x) = \frac{1}{1+e^{1-x}} \qquad (3)$$

Objective of using sigmoid function is to chart the predicted values with respective probabilities. The equation 3 denotes the sigmoid function.

RESULTS AND DISCUSSIONS

This section elucidates the results and discussions about the research carried out. After the vectorization, the data was splinted into two categories train and test, followed by the training the data, 30% of the original data was tested. The testing was carried out for different classifiers such as Gaussian Naive-Bayes(GNB), Logistic Regression(LR), Linear Support Vector Classifier(LSVC), Random Forest Classifiers'(RFC) as mentioned in the figure 6 and figure 7, in which Linear support vector classifier and Logistic regression yielded the overhead accuracy of 80.98% and 79.93% respectively as tabulated in the table 1. The Bengaluru traffic data collected from twitter was yielding to 80.98% was the contribution from our research work.

CONCLUSION

In this Research paper, the tweets are processed and different machine learning classifiers like naive bayes, are applied to find the sarcasm in the tweets, which in turn increases the over efficiency of the sentiment classifications by increasing the accuracy of the classifications. Gaussian Naive-Bayes, Logistic Regression, Linear Support Vector Classifier, Random Forest Classifiers' are used for testing the given texts, out of which Linear Support Vector Classifier and Logistic Regression had produced the better results comparing to other classifiers. Though Naive-Bayes produced good results earlier for sentiment classification, LSVC produces better results for determining the tweets with regard to sarcastic. The accuracy yielded by naive bayes was the least. It is overriding to note that though naive bayes performed better in the lexicon based models compared to machine learning approach.

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Table 1: Results of classifiers

Classifiers	Accuracy
LSVC	80.98%
LR	79.93%
RFC	77.38%
GNB	59.97%
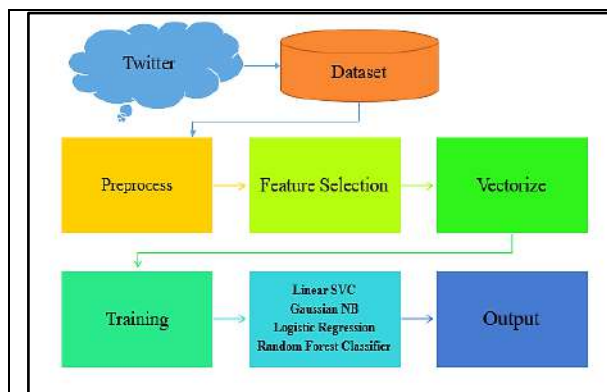
| Figure 1: Data Flow diagram | Figure 2: Sample sarcasm tweet |
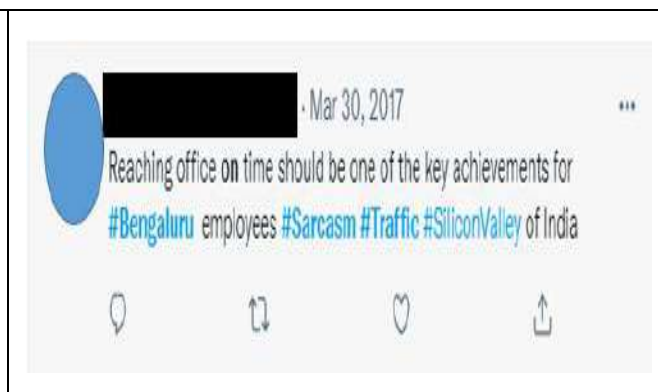
40800
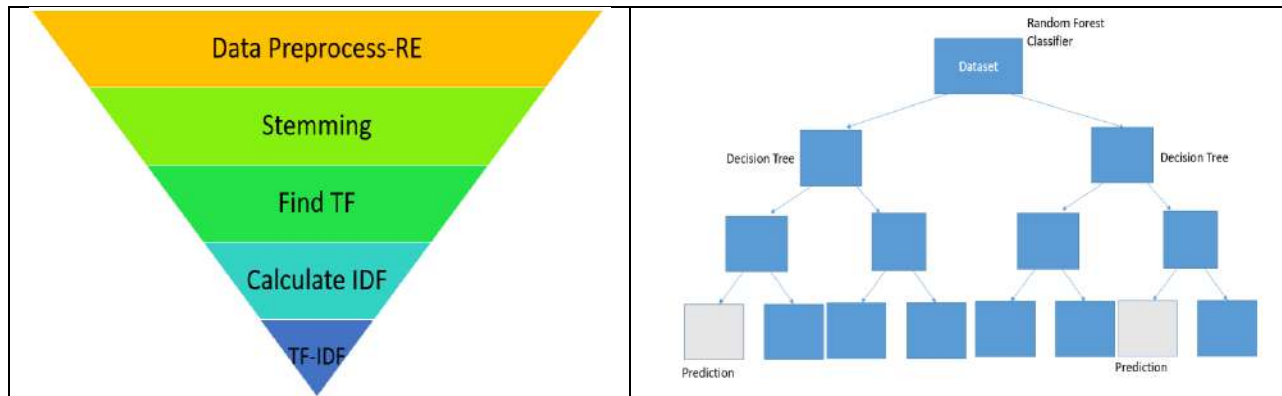
Figure 3: TF-IDF

Figure 4: Random forest classifier

Figure 5: Sampling of Logistic Regression model

Figure 6: Computation of classifiers
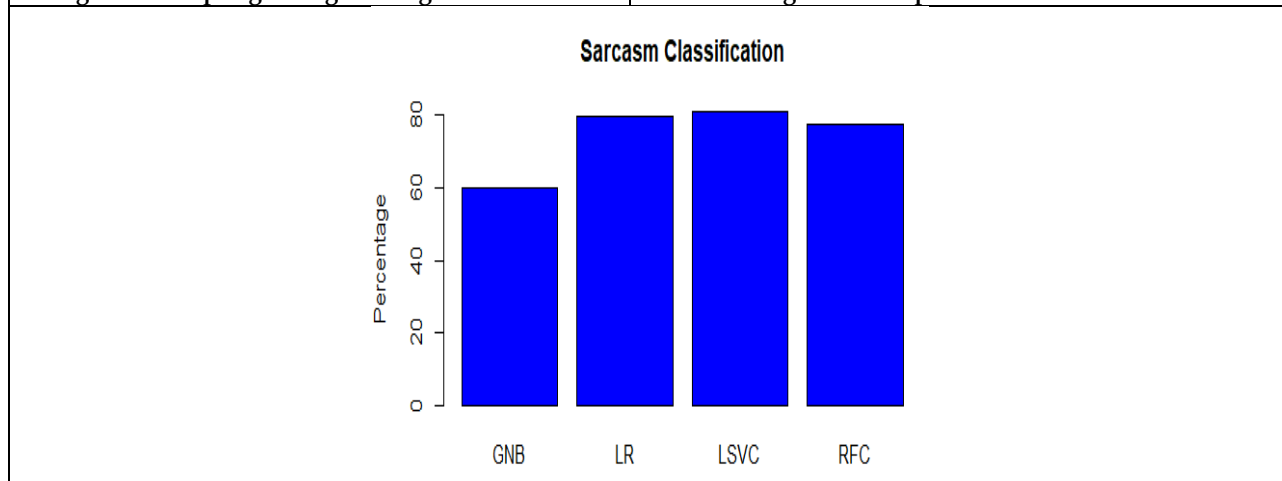
Figure 7: Sarcasm Classification

RESEARCH ARTICLE

Public Perception on the Performance of Local Government Employees

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ABSTRACT

Basic services provided by local governments are pivotal for the development of urban dwellers. Local governments offer a number of services to inhabitants that have direct influence on their lives. Since last few decades the role of local administration has changed swiftly across the globe. The enhanced pressure from different interest groups for citizen – centrism has prompted the essentiality to introduce marketing and management philosophies that depict the private sector. Comparatively a few studies have emphasized on the evolving trends of development in surpassing the bureaucratic paradigm in local government. The contemporary study examines and explores the citizens' satisfaction with the performance of employees in the local government of Visakhapatnam City in Andhra Pradesh, India. The study has followed an exploratory approach and a well-structured questionnaire has been distributed to 480 residents under the city jurisdiction. The results of this study probably help the local governments to recognize the gaps in employee performance and to take measures to enhance their performance.

Keywords: Customer Satisfaction, Local government, Employee Performance, Greater Visakhapatnam Municipal Corporation.

Introduction

The improved communication and developed technologies have enhanced the citizens expectations on services from the local government. Improved communication tools provided opportunity to citizens to compare services offered by various municipalities. The urban local bodies are required to put more efforts in order to encounter the expectations of their citizens.The pressure from citizens and various interest groups has led the urban local bodies to change their role. The enhanced pressure triggered the municipal corporations to implement management and marketing strategies that are part of the business sector. Inadequate financial resources is one of the important challenges most of the developing nations are facing in provision of basic services (Rao, 2020). Urban local bodies work to satisfying their inhabitants through implementing excellence tests and satisfaction surveys (Deniz Akgul, 2012). The customer satisfaction research in a local government context help to get insight into customer knowledge levels about the services provided, to identify priorities for services enhancement and to monitor the changes in expectations and performance. (Richard Boyle 2020). If the inhabitants are gratified with the performance of

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employees in local government, they may render more support to the system. If the local bodies are unable to gratify the demands of the citizens, they are likely to express their dissatisfaction with the local government and also with the democratic setup in the entire nation (Weitz-Shapiro 2008).In most of the European countries the public sector has adopted the corporate philosophy that assumes the marketing tools and strategies (Cousins, 1990). The principles of modern public management have brought a larger degree of decentralization and active involvement of citizens (Livingstone, Lunt and Miller, 2007). The inhabitants use municipal services and subsequently they ought to be in a situation to select the service providers depending on the quality (King, 2007).

The new public management has focused on citizens involvement in the process of public policies making(Cassia and Magno, 2009). The main aim of customer concerned business is to develop a long-term association with customers. The customer-oriented corporates deliver superior satisfaction to the customer and they have the capability to outrun the non-customer concerned enterprises (Narver and Slater, 1990). The customer orientation notion was developed initially for the business sector, whereas, some experts recommend application to other services (Lee et al., 2010). Numerous studies have been steered to explore the inhabitants changing role in public administration and enhancement in the public services delivery quality (Cicvarić et al., 2009). Whereas, a few studies have emphasized on the customer satisfaction with the local administration. The present-day study has been conducted to explore the civic opinion on performance of the employees of GVMC.

Objectives
1. To identify the efforts made by the municipal corporation employees in delivering basic services.
2. Study the public perception on the performance oflocal government employees
3. To offer basis for development needed to be accomplished within the local administration

Research hypotheses
Hypotheses formulated for the study are:
HO1: The public is not contented with theperformance of the local government employees.
HO2: There is a significant discrepancy inresidents' opinion (between zones) on the performance of employees of the local government.

Research Methodology

The study was grounded on both primary and second data.A structured paper-based questionnaire has been implemented for collection of primary data. The sample size was 480 persons. The sampling method was stratified random technique. With respect to gender issues, out of the total samples 50% were female. The major sources of secondary data were the reports, research papers, office of the municipal corporation and various websites. SPSS software package has been utilized for analysis of data. A one-way analysis is coxed among the groups to assess the inhabitants' opinion in different geographic zones under the jurisdiction of the municipal corporation.

Profile of the study area
The Visakhapatnam city is one among the fastest growing cities in India and the financial capital of Andhra Pradesh. The city was a tiny fishing village and later expanded with the development of port and establishment of industries. The public of nearby districts and different states visit the city for education, health and employment. The population of the city has increased swiftly and also expanded geographically by merging the nearby villages. The city of Visakhapatnam has been divided into geographic zones for the purpose of administration. The current study has been carried out in six geographic zones under the municipal corporation.

Organisation of GVMC
The municipal corporation is controlled by two divisions- deliberative and executive. The deliberative includes the General body theStanding Committee and Ward Committee. The deliberative wing has the role in formulating rules, regulations, policies and other administrative matters. The details of deliberative wing are depicted in the figure 1.

The executive division is headed by the Municipal Commissioner and he is supported by the Additional Commissioners, Chief Engineer, Chief City Planner, Examiner of Accounts and Project Directors reporting under the Commissioner. The organogram of GVMC executive wing is depicted the figure 2.

Public opinion on employees of the municipal corporation

The following portion depicts the opinion of the consumers on the performance of employees of the municipal corporation. The identified variables were subjected to factor analysis for determining the factors that are highly important for the consumers utilizing services provided by Greater Visakhapatnam Municipal Corporation.

The variables have been identified as important for the consumers utilising services provided by GVMC and are further considered for enhanced study based on the order of importance. The variable friendliness of municipal corporation officials occupies highest importance among the respondents with eigen value .969 followed by helpfulness of municipal corporation officials, easiness of identifying the relevant unit or person, time spent to get hold of the relevant person, the ability of municipal corporation to respond to complaints and the ability of municipal corporation to rectify problem. The Cronbach's Alpha score of the recognized variables is .954 which is extremelydependable.

Employees of municipal corporation should be approachable and table 4 portraits the opinion of the consumers on the friendliness of municipal officials. The overall mean score (3.31) specifies that the officials of municipal corporation are friendly. In comparison to other zones, the Zone 1 respondents are highly satisfied (with a mean score of 4.03) and Zone 5 people are least satisfied (with a mean score of 2.32).A one-way analysis of variance between groups is conducted to know the opinion of the consumers on the friendliness of municipal officials in different zones. There is statistically significant difference at the $p < .000$ level in the scores of friendliness of municipal officials for six zone groups at F (33.836). Despite reaching statistical significance, the actual difference in mean scores between groups is insignificant. There is no statistically significant difference in mean scores between GVMC Zones.

Table 5describes the helpfulness of GVMC officials and the overall mean score indicates that the municipal officials are helpful. With a mean score of 4.00 the Zone -1 respondents are satisfied more and the Zone 5 public are least satisfied with the variable helpfulness of municipal officials. The opinion of the consumers on the easiness in identifying the concerned unit or person to contact is portrayed in table 6. The overall mean score (3.45) clearly points out that it is easy to identify the relevant person to contact. The public of Zone 3 are more satisfied and Zone 5 are least satisfied with this variable.Table 7 represents the consumers opinion on the time spent to get hold of the relevant unit or person. The overall mean score (3.45) points out that the time spent to get hold of the relevant unit or person is comfortable. As per this table Zone 3 people are more satisfied (4.02) and Zone 5 are less satisfied (mean score 2.29) with this variable.

Public complaints should be addressed efficiently and table 8 explains the opinion of the consumers on the ability of municipality to respond to complaints. The overall mean score (3.03) specifies that the municipal corporation is able to respond to public complaints. Table 9 represents the opinion of the consumers on the ability of municipal corporation to rectify problems. The overall mean score (3.03) specify that the municipal corporation is able to rectify problems. The results of the study decisivelyreject the null hypothesis that there is anextensive disparity in thepublic opinion ofvarious geographic zones on the performance of employees of the local government.

Conclusion and Suggestions

The current study emphasized on the community perception on the performance of employees of the GVMC. The study outcomediscoversthat the mean score of the variables associated with community perception on employee performance is 3.26 on a 5-point scale. The mean score of the identified variables related to employee performance exemplify that the inhabitants have optimistic opinion towards the performance of municipal corporation employees. Whereas, the public is not completely satisfied with the employees' performance. This suggests that GVMC need to fill the gap in improving the performance of employees.Recruitment of employees would be made in time as per the requirement. Training programmesfor employees should be arranged periodically to adapt to modern technology and for providing civic services to the citizens more efficiently. Both hard and soft skills to be included in the employees training programs. Periodical analysis and reporting on local administration activities and tracking of citizens satisfaction are suggested. Recognition of performance and punishment to non- performers to be introduced to motivate the employees. Investment in employees' capacity building and a holistic approach are suggested for further enhancement of employee performance.

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Table 1: Factor analysis of the identified variables

Rotated Component Matrix

Variables	Component				
	1	2	3	4	5
1. Friendliness of mc officials	.969				.001
2. Helpfulness of mc officials	.948			.127	
3. Easiness of identifying the relevant unit or person to contact	.935		.135		
4. Time spent to get hold or the relevant person	.932		.160		
5. The ability of mc to respond complaints	.896	.440			
6. The ability of mc to rectify problem	.896	.440			

Table 2: Order of importance of the variables

Order of Importance	
Variables	Eigen value
1. Friendliness of municipal corporation officials	.969
2. Helpfulness of municipal corporation officials	.948
3. Easiness of identifying the relevant unit or person to contact	.935
4. Time spent to get hold of the relevant person	.932
5. The ability of municipal corporation to respond complaints	.896
6. The ability of municipal corporation to rectify problem	.896

Table 3: Mean value and Standard Deviation of recognized variables

Variables	Mean	Standard Deviation
Friendliness of Municipal Officials?	3.31	1.025
The helpfulness of municipal officials?	3.30	1.060
Easiness of identifying the relevant unit or person to contact?	3.45	1.029
The time spent to get hold of the relevant unit/ person?	3.45	1.041
The ability of the municipality to respond to complaints?	3.03	1.150
The ability of municipality to rectify problem?	3.03	1.150

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Table 4: Friendliness of Municipal Officials

Zone	Weighted Score	% to MPS	Mean	F
1	149	80.54	4.03	
2	76	63.33	3.17	
3	180	78.26	3.91	
4	129	78.18	3.91	33.836**
5	137	46.44	2.32	
6	174	62.14	3.11	
Total	845	66.27	3.31	

**p<.01 and *p<.05

Table 5: The helpfulness of Municipal officials

Zone	Weighted Score	% to MPS	Mean	F
1	148	80.00	4.00	
2	89	74.17	3.71	
3	174	75.65	3.78	
4	129	78.18	3.91	32.084**
5	135	45.76	2.29	
6	166	59.29	2.96	
Total	841	65.96	3.30	

**p<.01 and *p<.05

Table 6: Easiness of identifying the relevant unit/person to contact

Zone	Weighted Score	% to MPS	Mean	F
1	148	80.00	4.00	
2	96	80.00	4.00	
3	185	80.43	4.02	
4	129	78.18	3.91	39.299**
5	136	46.10	2.31	
6	186	66.43	3.32	
Total	880	69.02	3.45	

**p<.01 and *p<.05

Table 7: Time spent to get hold of the relevant unit/person

Zone	Weighted Score	% to MPS	Mean	F
1	148	80.00	4.00	
2	96	80.00	4.00	
3	185	80.43	4.02	
4	129	78.18	3.91	38.965**
5	135	45.76	2.29	
6	187	66.79	3.34	
Total	880	69.02	3.45	

**p<.01 and *p<.05

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Table 8: Ability of municipality to respond to complaints

Zone	Weighted Score	% to MPS	Mean	F
1	138	74.59	3.73	
2	56	46.67	2.33	
3	157	68.26	3.41	
4	127	76.97	3.85	18.991**
5	132	44.75	2.24	
6	162	57.86	2.89	
Total	772	60.55	3.03	

**p<.01 and *p<.05

Table 9: Ability of municipality to rectify problem

Zone	Weighted Score	% to MPS	Mean	F
1	138	74.59	3.73	
2	56	46.67	2.33	
3	157	68.26	3.41	
4	127	76.97	3.85	18.991**
5	132	44.75	2.24	
6	162	57.86	2.89	
Total	772	60.55	3.03	

**p<.01 and *p<.05

Source: Office of the Additional Commissioner (Gen), GVMC

Figure 1: Organogram – GVMC Deliberative Wing
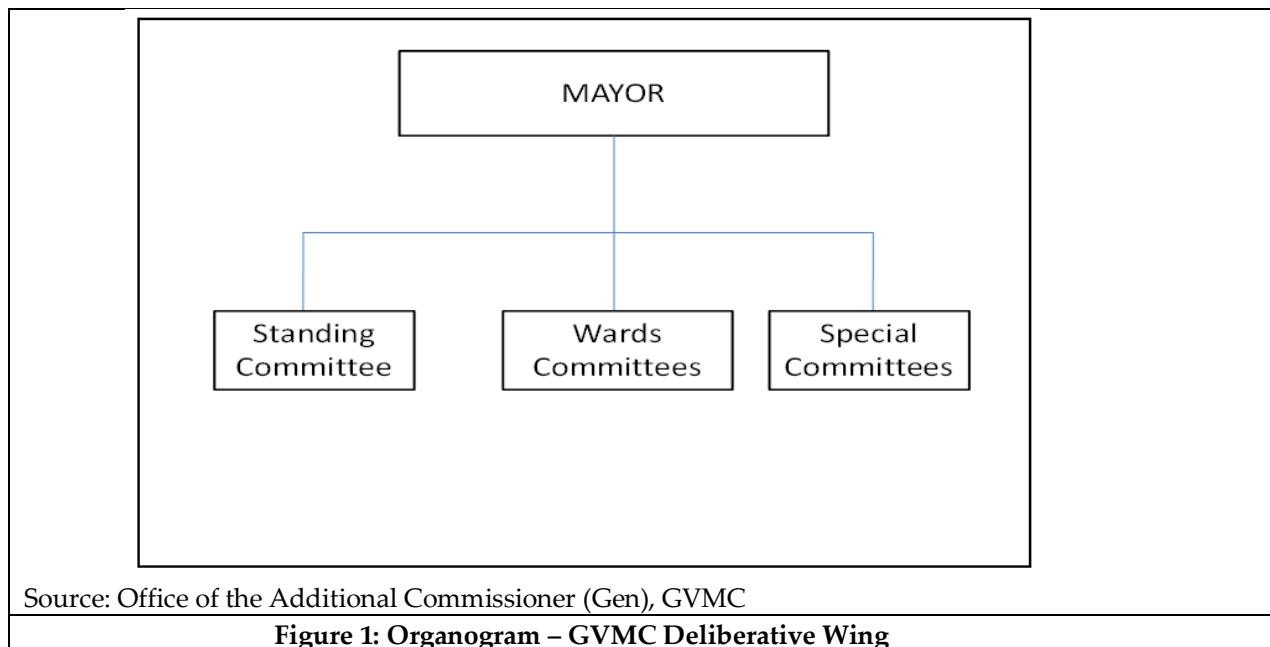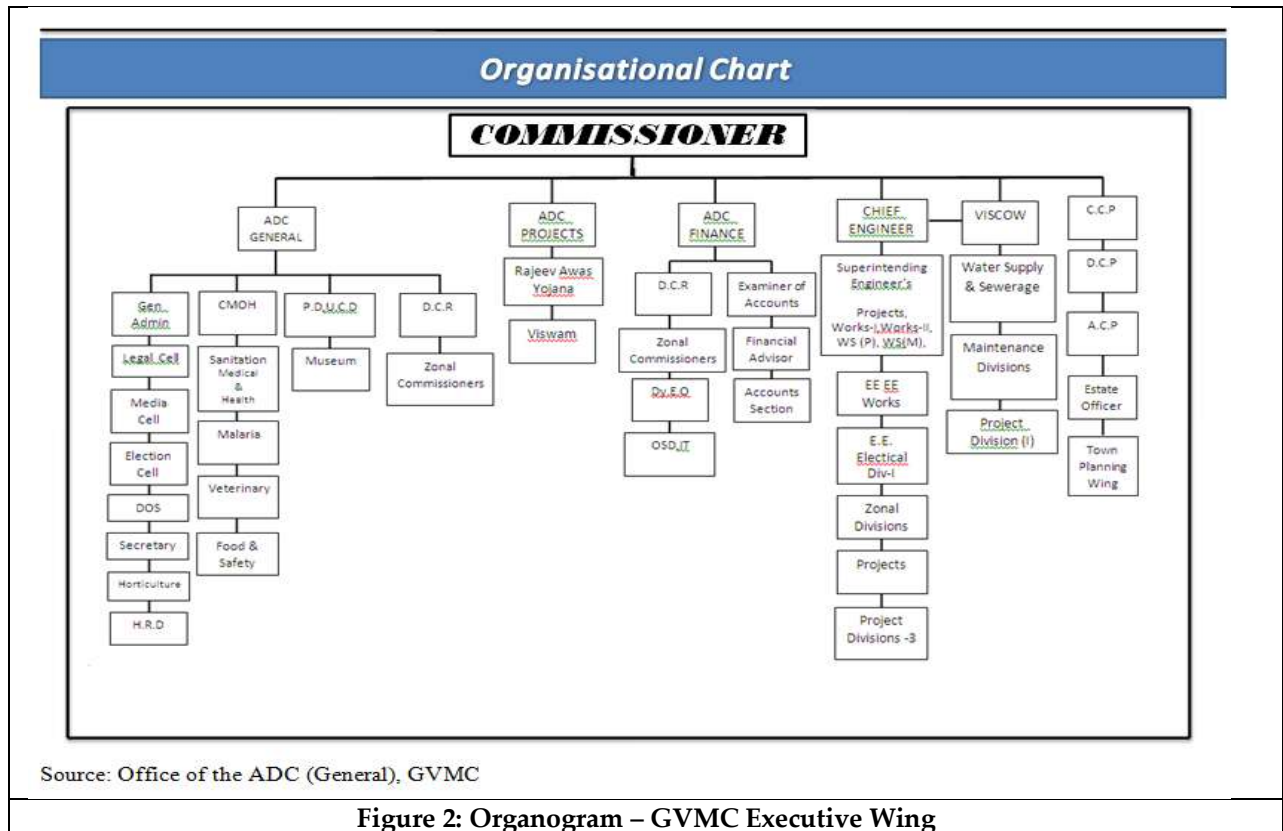
Figure 2: Organogram – GVMC Executive Wing

RESEARCH ARTICLE

Comparing Factors Responsible for the Top Performance of a Student in Different Academic Fields

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ABSTRACT

The objective of this study is to determine what are the most essential characteristics and environmental aspects of a top performer. This research also aids us in comparing variables that have a significant impact on their performance. Science, mathematics, and reading are among the academic fields under consideration. The dataset used in this work is collected from PISA 2018 secondary data science scores, with top performers having proficiency levels 5 or 6 from a total of 6 competence levels. We use different algorithms, specifically SVM - RFE-linear (Support Machine Vector, with linear Recursive Feature Elimination Kernel), Random Forest, and a hybrid model SVM-ANN. We use SVMRFE and Random Forest to rank the top features and use all 3 models to predict the performance based on these selected features. Here the SVM-ANN outperforms Random forest and only slightly outperforms SVM-RFE. The metrics used for result comparison are accuracy and recall. SVM-ANN provides a better outcome with accuracy values of 0.8185, 0.8383, and 0.8053, whereas random forest provides a good recall value of 0.8648, 0.8648, and 0.8644 for reading, science and mathematics respectively.

Keywords: Machine Learning, Deep learning, Student's performance, Support Vector Machine, Recursive Feature Elimination, Random Forest, Artificial Neural Network.

INTRODUCTION

The study of science and mathematics has always been crucial for any developing/developed nations/economies and as for reading it helps in developing critical thinking skills as well as better comprehension. These are critical for technological advancement in any industry. So, aside from a natural liking for the subjects, it is critical to grasp what distinguishes a top performer from the rest. Programme for the International Student Assessment (PISA) is a programme developed by OECD for the assessment of various academic skills of middle school students. This program measures the expertise of students in different areas of academics such as Maths, scientific literacy, and reading capability and condenses them into numeric values called Plausible Values (for example PV1SCIE - this

stands for Plausible Value 1 for scientific literacy). The PV system already accounts for differences in different economies (nations) and conducts the tests accordingly. This gives a global measurement of students' abilities. The dataset includes school-level characteristics such as the availability of IT, the quality of labs, the interactiveness of teachers, and so on. It is made up of data from 6,00,000 pupils in the schools of 79 participating nations and economies.

Literature Review

To get more insights on the problem statement various papers were referred which have been further summarised below: Chen J. et al[1] attempts to identify the distinguishing features between the top performers and non-top performers in science literacy of students in secondary schools at varying levels with the use of SVM-RFE and CART. SVMRFE with 77.5% accuracy (20 feature model) was chosen over CART for its reliability over CART. The survey used to collect the data was descriptive in nature and the survey is filled out by students themselves. Gorostiaga et al[2] presents a simple and flexible feature extraction approach that provides the relevance level of the characteristics' impact on the desired variable. A total of four techniques are used in the analysis of their data, with FDA, SVM Linear, and SVM non-linear coming on top. To reach more generalizable results, it is necessary to carry out new studies, analysing consecutive years.

Beaulac et al[3] creates two classifiers using random forests in their study of predicting academic field and performance of the students. With 200 trees, the random forest produced with the R Random Forest package had the best cumulative accuracy of 78.84 percent. Computationally more intensive than classical RFE-Guyon, which makes it hard to implement on huge data. Sanzana et al[4] attempts to define and characterise Chilean student characteristics based on mathematics academic performance. Statistical approaches such as random forest coupled with classification and regression trees were employed, with individual and family behaviour data. The CART was a better model according to RMSE. The high computational load of both SVM techniques is a drawback. Sanz et al[5] use SVM_RFE to filter and visualise only the crucial variables with the help of non-linear kernels, by using both the linear and non-linear versions of SVM, three approaches to rank variables are proposed. Even though usually the RFE-pseudo-samples performs better than the alternatives, it requires a large amount of computational power when compared to the classic SVM-RFE. This would be a problem in bigger datasets, and datasets with missing values. Alivernini[6] compares 20 different countries to find common elements in both extreme low and high performing students in Progress in International Reading Literacy Study (PIRLS). The method used was classification and regression trees. The findings demonstrate that the teachers with more than a decade and half of experience coupled with change in their pay is what makes the most difference in terms of pupil level of success.

Agasisti et al[7] aims to figure out what qualities are shared by top performers with a socio-economically challenged background status and attend underprivileged schools. The authors determined that individual level features, as well as some school elements, play an equal influence in developing a top performance after conducting a multilevel logistic analysis. The results obtained are specifically obtained from the data of Italian students, so it may be difficult to generalise it. Acosta et al[8] provides a study on the influence of parents' enthusiasm regarding science perception in adolescents in Hong Kong.The study considered guardians' SES [Students' socioeconomic status] and their attitude towards science. The other differentiating factors such as, students' gender, students' age, parents' occupation, school size etc were controlled for.   Chakraborty et al[9] provides a hybrid model of CTANN (Classification Trees - ANN) in the context of estimating student performance in academics. This model outperforms the conventional supervised learning models.

Preliminary

The goal of this study was to obtain the most impactful characteristics from the entire set of 942 variables that are required to characterise top performers from this big collection of possible features. We use a base model of SVM-RFE. Furthermore SVM-ANN a hybrid algorithm is utilised.

We attempt to identify and compare the most critical components for a top performer in science, mathematics, and reading comprehension at the student, family, and school level in this research. We will be utilising this PISA dataset

for our study. Firstly, we drop the features with more than 50% missing values and impute the rest using KNN. Then we perform feature selection using SVM-RFE and Random Forest. We then analyse this dataset using SVM - RFE (Support Machine Vector, with Recursive Feature Elimination), Random Forest, and a hybrid model of SVM-ANN.

Algorithms
SVM-RFE

The support vector machine is used commonly because it achieves excellent accuracy while requiring little computational resources (Fabio Alivernini 2013) (Sanz et al. 2018.). The purpose of the SVM approach is so that a border between the two classes can be obtained in a multidimensional space that categorises the data points clearly. Our space is mapped to a higher dimension for non-linear data. Borders created to divide the space are called hyperplanes. Hyperplanes are decision boundaries that help in data classification. These hyperplanes are then used to divide the space into chunks of space, with each chunk belonging to a particular class, and any data point falling under that chunk will be classified into the respective class of the chunk. Our goal is to draw the borders in such a way that the distance between chunks of different classes are maximised. Meaning we have to obtain the borders in such a way that the buffer between the classes are maximised. Making these margin distances (buffers) as large as possible makes it easier to categorise subsequent data points by giving ourselves some buffer.

Formula
Training dataset of n points of the form:

$$(x_1,y_1),...,(x_n,y_n) \tag{1}$$
$$y_i(w^T x_i - b) >= 1, \text{ for all } 1 <= i <= n \tag{2}$$

where, x_i is a p-dimensional real vector w is the (not necessarily normalised) normal vecor to the hyperplane

Random Forest

The Random Forest technique consists of an ensemble of decision trees, each of which is made up of a data sample selected from a bootstrap sample with replacement for the training set (Beaulac C., Rosenthal, J.S., 2019) (Beaulac C., Rosenthal, J.S., 2019.). The Random Subspace Method randomises which features are included. This implies that each tree goes through a training procedure, but since it has access to various characteristics, it will reach different conclusions. Along with it, randomization is then injected into the dataset using feature bagging, increasing variety and decreasing correlation among decision trees. It captures the fluctuation of numerous input variables concurrently and enables a high amount of observations to contribute to the prediction. A majority vote—i.e. the most frequent categorical variable—will produce the anticipated class in a classification problem. Finally, the oob sample is utilised for cross-validation, which completes the prediction.

Formula:
Here, pi represents the relative frequency of the class you are observing in the dataset and c represents the number of classes.
$\sum$ from i=1 to c

$$Gini = 1 - \sum(p_i)^2 \tag{3}$$

$$Entropy = \sum -p_i * \log_2(p_i) \tag{4}$$

The branching of nodes is based on entropy. As given above the entropy utilises the chance of certain outcomes occurrings.

SVM-ANN

The result from SVM was taken as an additional variable for ANN, thus creating a hybrid model (Chakraborty T. et al., 2018). Artificial neural network as the name suggests which is built to mimic a collection of biological neurons, is a computational model that has the ability to perform a range of tasks such as classification, decision making etc. The

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layers of this network can be divided into 3 parts, the input layer, the output layer, and the hidden layer. Each layer consists of nodes, these nodes are called neurons. The input values are entered into the input layer. Then the deep learning process happens in the hidden layer, which is often called black box, as we are unaware of the processes happening within this hidden layer. The hidden layer will consist of one or more neural layers. Then the output of the processes that occurred within the hidden layer is output onto the output layer. The connection of neurons between layers and within layers is called a weight, which is represented by a numerical value. The neural network uses an activation function to deal with nonlinearity present in the data. As the problem at hand is a binary classification we use the sigmoid function as our activation function.

$Y = (input*weight) + bias$ (5)

$z = x_1*w_1 + x_2*w_2 + \ldots + x_n*w_{n+b}*1$ (6)

$y = a_{output} = sigmoid\ (z)$ (7)

$sigmoid(z) = 1/(1+e^{-z})$ (8)

Here, x_i = input   and w_i = weight

METHODOLOGY

Dataset

The data set (https://www.oecd.org/pisa/data/2018database) is a survey that was participated in by middle school students. The data was then compiled and put together in a usable format by the PISA under OECD. There were mainly two questionnaires that needed to be filled. One was for the student, and the second had to be done by the principal of the respective school. There were more surveys that could be filled by the parents of said students. But seeing as these surveys were optional. The extra information obtained by these additional surveys were kept to minimum, meaning most of the information obtained by these extra surveys, can be obtained through the two main surveys. Seeing as such the main two surveys give us the most information in any case, and the addition of these extra surveys in our dataset would only result in us dealing with more missing values, only the main surveys were chosen. The performance of students in various fields of academics were indicated using a set of categorical variables, from a score of 1 to 6. Students at the level of 5 or higher are considered top performers the rest are considered non-top performers. These values that grade a student's performance are derived from another set of variables called plausible values or PVs for short. These values would give an estimate of the student proficiency in a subject field. These estimates would be done multiple times (10) throughout the academic year, as to gauge the proficiency in the subject better. The 10 PVs are virtually indistinguishable, when considering large samples. Thus, a median value of PV was taken into account for each subject for each student. The features present in the dataset are either the original responses by the students/schools or they are derived from these responses by the OECD team. To know more about these features, please refer to the PISA-D Technical Report of 2018 put out by OECD.

Process

The data set was merged with the primary key being the student id. Variables with more than 50% of missing values were then dropped. Missing value treatment was done using two methods. Firstly, for variables common for students among schools was imputed using responses filled in by the students of that particular school. Secondly, the other variables were then imputed using KNN at the country level. Then the data was normalized using the min-max scaling method. A multi-step approach was then adopted to analyse the differences in top-performing students and the rest, and to find the differences in characteristics of top performers in different fields of studies. Specifically, science, mathematics, and reading comprehension. Firstly, random under-sampling was performed to address the class imbalance issue present in the dataset. Then SVM-RFE (Support Machine Vector with Recursive Feature Elimination Kernel) was used to obtain top 10 features for each of science literacy, mathematics, and reading comprehension. Then random forest, SVM-RFE classifier, linear regression, and SVMANN were implemented and feature lists and predictions were obtained for each field. To ascertain the performance ability of various models used, a cross validation of ten folds was used. The ten-fold cross validation divides the entire data set into 10 subsets

and proceeds to implement the model ten times, with every subset being the test dataset exactly once and the other nine being the train data set. The accuracy and recall were used as the measures of performance for the model.

Accuracy = Total number of correct classification / Total
number of Observations   (9)
Recall = Total number of top performers classified as such /
Total number of top-performers   (10)

The descriptive statistics for the important features were obtained to give detailed insights into the effect of these features on the performance on various fields of study. All the above mentioned processes including preprocessing, computing, and analysis were all performed on an open source software of python 3.7 in combination with additional packages imported from various open source documentation libraries from pystatread, pandas, numpy, scikit-learn, imblearn, and keras.

RESULTS

The features are eliminated one-by-one starting from the least important feature, which is determined by the coefficient assigned by SVM-RFE. That eventually gives an ordered least based on the importance of the feature in determining the performance of a student in various academic fields. We have done the above for science, mathematics and reading and selected 10 features for each field. Some of the characteristics were common for all three fields. This demonstrates how significant these characteristics are for any top performer, regardless of academic subject. Did they work for money before going to school, their environmental awareness, what are their Collaboration and teamwork, what is their desired education level, how many books do they have at home, and how well do they understand the environmental issues? The increase in greenhouse gases in the atmosphere were among the most significant aspects in all three fields.

Model Comparison

From the used methodologies, accuracy and recall from each model for science, mathematics and reading is given below in the table: Random Forest, SVM-RFE, and SVM-ANN were utilized for modelling purposes. In classifying top performers and the rest of the students, SVM-ANN slightly outperforms both SVM-RFE and Random Forest.

CONCLUSION

The objective of the paper was to determine a few of the crucial characteristics and environmental aspects of the high performing middle school students in various fields of academics. We can say 'Number of science classes in a week', 'Internet usage outside school for studies', 'Availability of digital devices in school' are important factors for performance in science. Whereas 'Number of mathematics classes in a week', 'Internet usage outside school for studies', 'Availability of Resources (Non- Internet)' are important factors for performance in mathematics. 'Student Motivation', 'Availability of digital devices in school', and 'Family wealth' are important for developing a reading comprehension in students. As for the models used,. SVM-ANN loses explainability. SVM-RFE and Random Forest, on the other hand, maintain their explainability. We may utilize decision trees to better understand how variables affect top performers in terms of whether they affect them positively or adversely, and how much they affect them. While doing knn imputation we can also try using language as filter instead of country to see how factors affecting top-performers change.

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Table 1: Top 10 features for each stream

SI.No	Science	Mathematics	Reading
1	Number of science classes in a week	Number of Mathematics classes in a week	Student Motivation
2	Internet outside school usage for studies	Internet usage outside school for studies.	Usage of computer
3	Availability of digital devices in school.	Interest in physics concepts.	Availability of digital devices in school.
4	Self-efficacy in Science	Parents emotional support	Family wealth
5	Highest Education of parents	Availability of Resources (Non-Internet)	Availability of Resources (Non-Internet)
6	Usage of internet devices for nonstudies.	Student Motivation	Contact with teachers outside school.
7	Environmental Awareness	Do they install new software on their own	Handling of feedback.
8	Availability of teachers.	Collaboration and teamwork	Collaboration and teamwork
9	Teacher support	Student Attitudes, Preferences	Personality: Test Anxiety
10	Do they have concerns about receiving poor grades at school?	When learning school science students are asked to do an investigation to test ideas	Interest in broad science topics.

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Table 2: Result comparison

	SVM-RFE		Random Forest		SVM-ANN	
	Recall	Accuracy	Recall	Accuracy	Recall	Accuracy
Reading	0.8484	0.7903	0.8648	0.7765	0.8102	0.8185
Science	0.8566	0.7948	0.8648	0.7732	0.8104	0.8383
Maths	0.8557	0.7672	0.8644	0.7609	0.8057	0.8053
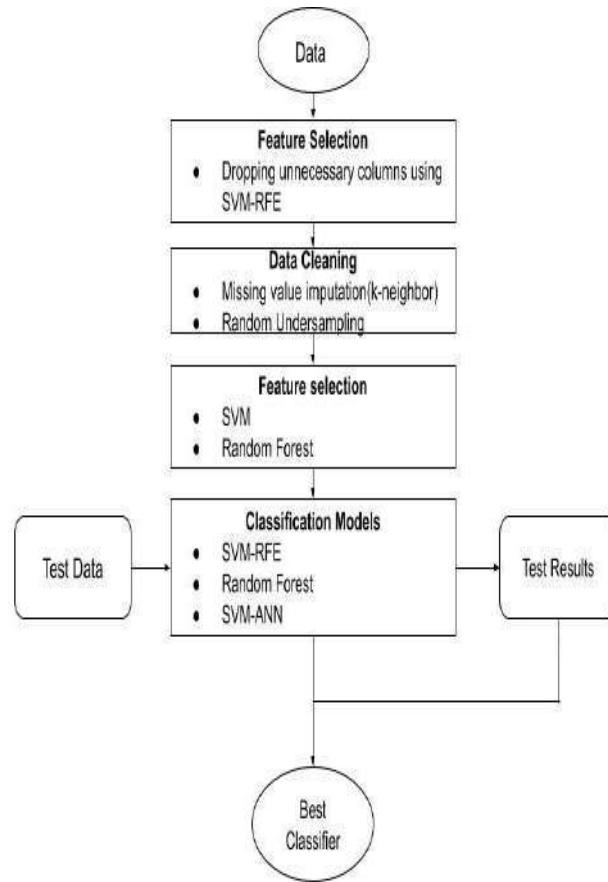
Figure 1. Proposed Work flow

RESEARCH ARTICLE

A Survey on Elephant - Train Collision in India

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ABSTRACT

The collision between human and animal will remain in its vein, but as per the ecological concept of existence , humans alone cannot survive as a single species; they need to have an ecological balanced environment . In this survey paper , the conflict between humans and elephants has been focused in the form of accidents in the Indian dense forest area, especially near the railway tracks . The colliding factors have been found in an increasing graph due to some natural or artificial reasons. To mitigate the colliding reasons some sort of technological spare have been utilised and projected .But still the survey finds no concrete or permanent solutions have been used to evaporate the problem .

Keywords: Animal-Movement, Sensors, Internet of Things, Image Processing, elephant-train collision, Convolution Neural Network(CNN).

INTRODUCTION

The biological forest diversity of India covers an area of 721,249 square kilometre approximately[which is equivalent to 21.67% of India's entire geographical area. The biota of forest attracts a wide range of animal to have it as the habitat. Elephas maximus or Asian Elephant are considered as the only living species which belongs to genus Elephas and it is collectively found throughout the Indian Sub-continent along with South East Asian regions of India, Nepal Sumatra and Borneo in west ,north, south& east respectively. Elephant is one of the keystone species of Indian forest which plays an important role in maintaining the biodiversity of the area which they live. According to a popular e newspaper DTNEXT statement on Jan09, 2019 , India accounts for over 50% of elephants population of Asia with an estimated number of 27,312.At the same time this number is reducing due to some biological challenges which indirectly force the herd to come out of the forestial territory and collide with the unexpected probability of death .It has been observed that the unnatural accidental death of elephants are mostly due to collision with train. According to Wildlife Trust of India, from 1987 to 2018, around 281 Elephants are dead from train accidents only as because 20 of 101 elephant corridors are intersected by railway tracks[35].

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The most train accidental death incidents of elephants have been observed at night[1][2] during the two time spare. One during the months of high temperature with low low rainfall areas and other during the time of crops raiding. The death ratio of male elephants has been found the most with respect to females. Due to scarcity of water in the forest regions during the summer time necessitate the elephants to transpire of their dwellings and similarly due to deficiency of food availability ,forces them to move towards the crop raiding villages. Now because of both the reason they incidentally meet with the train collision which interrupts on their path. A deep survey has been conducted to find out several hotspots(the most used path by the Elephants near railway tracks) to mitigate the number of accidents especially during night. As Elephant conservation has now become a challenge so to mitigate the train-elephant accident , lots of updated technological aspects along with elephant detection algorithms have been proposed and still some are on projectile.

Observed Facts

Starting from the year 1987 it has been observed that there are 101 Elephant Corridors in India among them 20 are Rail tracks crossing, that allow them to move between habitats[35]. Some of the following notable reasons that leds to train-elephant collision have been observed :

a.   During the month of high temperature and low rainfall month the species have to face the water scarcity problem which led them to move to

nearest populated area for survival.[1][20][15]

b.   The movement has been observed during night and early morning.[1][9][20]

c.   Due to habitats modifications [20]

d.   The time of Crops raiding is one of the important fact observed which leds the elephant to come

out of their habitat .[1][20][15]

e.   A major artificial reason is been noted that during the early 90's the maximum speed for the metre gage has been fixed to 60 km/hr which is now developed to broad gage with a maximum speed

to 100km/hr .[2][20]

f.   A major accidental facts have been observed in the male species.[1]

Suggestions To Mitigate The Fact

The fact can be mitigated by the following suggested ideas

a)   Due to water scarcity in the summer and low rainfall , elephants seek water. So if habitats are been modified by developing recharging natural water resource especially during these months.[20]

b)   By reducing the train speed at dense forest area along with limiting train operations during the night time.[2]

c)   Fencing the track except the elephant common crossing corridor areas [2]

d)   To make an uninterrupted path by building of overpass and underpass near the dense forest areas to.[2]

e)   Implementation of artificial intelligence near the track to identify the object and broadcast the signal in the form of a message to slow down or to stop the train whichever is possible at an instant.

f)   To prevent the deforestation or illegal cutting of trees which disturb the ecological equilibrium of the forest and shortage of food for the wild animals. This will reduce the intervention of elephant in the crop land.

Statistical Figures

From the last few years some of the region of India has been noted for train-elephant collisions in a wide aspect. Some of them are mentioned below in a tabulated format. From the reports of various studies it has been discovered that as an average 80 numbers of elephants are killed every year. It has been counted out that between the year 2009-2010 and 2016-2017 ,an approximate number of 655 elephant deaths has been encountered with human conflict and among them 120 deaths has been found out due to collision with train .Elephant train accident cases in Assam are been recorded as the highest ,followed by West Bengal ,Uttarakhand, Jharkhand ,Tamil Nadu ,Uttar Pradesh, Kerala and Odisha with 26%. ,14%, 10% , 6%, 6% , 3% and 2% respectively. [30] 2016-2018[30][35]

As per the 2017 report of Elephant Census , it has been counted out by the techniques of direct & indirect along with waterhole methods and the mapping method of elephant distribution, that the number of elephants has been found to get reduced by 3000 within the last 5 years.[31]

Technological Trials

On July 2015, A. K. Kulatunga, J. Gowrynathan, R. Ekanayake, D. Athauda, C.Chandrakumar , from the Department of Production Engineering, of University of Peradeniya, Peradeniya, Sri Lanka[5], respectively have collected the data as reasons of colloidal and categories them into three locomotive related problems, site related problems and driver related problems. To overcome with the locomotive related problem , a suggestive model tested at an temperature of -20°C to +60°C and non condensing humidity of 5% to 95% As major components, the model consists of 660 grams of light weighted cameras of 2x Electronic zoom capacity with Field view of 24° (H) x 18°(V) and Field of view 12° (H) x 9°(V) with 2X extender and both having lens of fixed focal length of 19 mm along with Spectral range and thermal sensitivity of 7.5 to 13.5 µm and <50 mK respectively and maintaining an image frequency of 8.3 Hz Pal / 7.5 Hz NTSC, and the entire image data processed on an algorithm of Image processing Digital Detail Enhancement (DDE) has been implemented on the roof head of the locomotive which will help to display the object at a distance of 0-600m on the mounted display board installed in the driver's cabin.[5]

January 2016, Rana Biswas et al. : have suggested a system to avoid wildlife accidents which consists of pyroelectric element like Passive Infra Red(PIR) sensor(detects body heat) ,Buzzer,GSM SIM300, Ultrasonic ranging sensor and Fresnel lens embedded on Arduino Uno will send an message to base station on which will forward to driver's android device connected through satellite communication.[13]. January 2016, Dr. M. Prabu ,Department of Computer Science and Engineering, Adhiyamaan College of Engineering, Dr. M. G. R. Nagar, Hosur, Tamil Nadu, India[7]: has suggested an efficient surveillance system which will intimate the forest authority through a short message service after processing the data by FastICA algorithm of elephant position grabbed by GPSposition near forest borders by using seismic sensors . January 2016, Dr. M. Prabu, Dr. D. Thilagavathy Department of Computer Science and Engineering, Adhiyamaan College of Engineering, India[9]: proposed a system , will work on the acoustic wave of Elephant captured by acoustics receptors and process them using Adaptic filter & Mel-Frequency Cepstra Coefficients (MFCC) over Matlab to make a match with the elephant sound pattern on return of which a short message will get triggered to forest officials ,station master and train operator so that the necessary action can be taken to avoid the collision.

July   2016, Chandra   Kumar   J*   and   Selvakumar A Department of Energy and Environmental Engineering, Saveetha University, Chennai[19] : suggested a combination system thermal image processing and acoustic techniques.the infrared detector will send the data accrued by FLIR thermal imaging camera ,to image processing circuit to get convert into an thermal image format to get displayed over LCD .As an action the microcontroller will automatically create the buzz sound with gunshot and braking the train at the same instance of time. February 2017, SACHIN UMESH   SHARMA AND DHARMESH J. SHAH, Department of Electronics and Communication, Gujarat Technological University, Ahmedabad, India [17] has proposed a low cost computer vision based system to avoid collision by detecting the animal before the impact. The system has been successfully tested with an accuracy of 82.5% .Once the animal is detected it will convert distance from pixel to metre and will be able to send an alert message to the vehicle driver running at a maximum speed of 35 km/hr. February 2017, V.Elakiya , G.Ashwini Devi and K.Sagadevan Department of Electronics and Communication Engineering, IFET College of Engineering, India[18] proposed a sensor based system which comprises of seismic sensor and vibration sensor embedded on AT89S52 microcontroller on detecting the elephant the Proteus simulator will create a buzz sound with activating the electric fences and send an alert message to the control room. February 2017 ,D.Aravind, S.Anupriya, A.T.Aarthi, C.Saravana Kumar2,, ECE Department, Valliammai Engineering College, India[24] have suggested a wildlife tracking system which comprises of a tracking device implanted on animal body and an alarm will be generated if the object tries to get out of the GPS defined zone .

February 2018, Pravin A. Dhulekar, Sanjay T. Gandhe, Ganesh R. Bagad ,Sudhanshu S. Dwivedi, Department of Electronics and Telecommunication Engineering, Sandip Institute of Technology and Research Centre, Nasik, India[21] suggested a vision based animal detection technique to increase the detection accuracy and decrease detection time by using feature descriptor (Histogram of Oriented Gradient) ,a visual descriptor(Local Binary Pattern) processed using a Smart Vector Machine algorithm and Adaptive Boosting Classifiers. March 2018, Sivaraman Karthikeyan ,Sri Sairam Engineering college ,J. Syed Ali ,Porika Venkatesh, Indian Institute of Technology Delhi,M. Vignesh,M. Prathik have proposed a system of PIR sensors and IT temperature sensors with PIC controller embedded on Arduino Rev 3 over a Proteus simulator which will sense the animal and send the sensed data to base station to process and scrutinised by comparing previously collected data set and then the plantation workers will be intimate .[28]. April 2018, Mr.Prashant K. Kulkarni, AkshaySuyash Bal, Yogesh Namdev Bandekar, Pratap Pradeep Amare, Sindhudurg ShikshanPrasarak Mandal's College of Engineering (SSPM COE) Kankavali[4], introduced a view of Elephant Intrusion Detection System by detecting the elephant heard using filtered(Fast ICA algorithm with spectrogram) seismic signal produced by the seismic sensor and once this signal is matched with the stored pattern of elephant ,immediately the authority will be intimated through Short Message Service for correct measures to drive the heard away.

April 2018, Rakesh Kumar Mandal and Dechen Doma Bhutia Department of Computer Science & Application University of North Bengal, Darjeeling, India[6], proposed an elephant detection model comprising of an Artificial Neural Network Model with weight detection algorithm and 4 Geophone sensors at a distance of 5km each near the railway tracks over a radius of 15 km,10 km and 5 km. June 2018, Jerline SheebhaAnni , Arun Kumar Sangaiah School of Computing Science and Engineering, VIT University, Vellore, Tamil Nadu, India[3], early warning systems near railway track by using Wireless sensor networks which includes acoustic sensor to detect elephant motion and pattern, vibration sensor for age and gender classification and a camera .This system will work on the principle of cognition theory and Fuzzy Cognitive Map. July 2018 ,Mr.A. ANANDHA RAJA1, Ms.K. RAMYA2, Ms.B. KOUSALYA3, Mr.S. JEEVA Assistant Professor, Department of ECE, SNS College of technology, Coimbatore, Tamil Nadu, India[12], suggested a system to prevent the wild animals from accidents by using the analyses technique over Passive InfraRed sensor(PIR) captured data by and processing on Matlab which will turn the traffic light/signal to red. March 2019, Omkar Vivek Sabnis and Lokesh Kumar R ,School of Computer Science and Engineering, Vellore Institute of Technology, Vellore, India[10]: suggested a nobel object detection system which will operate on the principle of object detection by implementing overhead cameras at the elephant crossing prone site and once the detected object found on the track or vicinity , an alert message to both the nearest station will be send to slow down the train. July 2019, N S GogulDev ,K S Sreenesh,P K Binu Amrita Vishwa Vidyapeetham Amritapuri, India[16] : suggested an IOT based automated system to detect the animal near crop land using PIR sensor and a camera with a TensorFlow image Processing technique which will create a buzz sound embedded on Raspberry PI.

September 2019, CT Wijewantha,WPJPremarathne and WMKS Ilmini ,Faculty of Computing, General Sir John Kotelawala Defence University, Ratmalana, Sri Lanka[22] : have introduced the concept of machine learning in order to reduce frequent killing of elephant from train collision. Introduced the application of drone to capture image and then depending on the motion ,skin colour and PIR sensor data , accuracy of Elephant detection will be reached and then as a response the alert message will be triggered using the radio frequency to either the user or the nearest railway station. April 2020, T. Thomas Leonid ,KCG College of Technology, Chennai, India & R. Jayaparvathy ,S S N College of Engineering, Chennai, India[25]: suggested a model with 93.32 % accuracy with respect to feature data dimension using Mel-Frequency Cepstral Coefficient (MFCC).The system comprises of feature extraction methods with Principal Component Analysis(PCA) and a Support vector machine to classify the object as elephant depending on the voice of elephant. July 2020, Bianca A. Ferreira Council for Scientific and Industrial Research, J. Pieter de Villiers University of Pretoria, Allan de Freitas University of Pretoria, South Africa[23] : have suggested a system to detect and classify the digital photographic image data using deep neural network over a network structure of YOLOv3 network with an accuracy of 86.64%. August 2020, Aritra Acharyya, PrasenjitDey, Abir Datta & Sukanya Bose: suggested an analytical model of video surveillance system which will be implemented along the railways tracks or roadways to determine the parameters like angular alignment, position, length of a proposed real time

video surveillance work for 24x7 in order to avoid the animal-train or animal-vehiclecollisions.[14] August 2020,Atri Saxena,Deepak Kumar Gupta,Samayveer Singh, Dr. B.R. Ambedkar National Institute of Technology Jalandhar Jalandhar,India[27] : proposed a system to detect the animal using SSD which is a single deep neural network and deep convolutional network of Faster R-CNN for faster data detections with that of previously suggested methods. January 2021, Sayan Dutta of IEM, Arati Paul & Debasish Chakraborty & G. Srinivasa Rao[37] of ISRO has proposed a model to detect the elephant using HAAR feature extraction technique and boosting based machine learning algorithm in a real time video data. November 2021, Surbhi Gupta, Neeraj Mohan, Padmalaya Nayak, Krishna ChythanyaNagaraju& Madhavi Karanam[36] proposed a deep learning model based on vision ,automated and warning system to detect as well to identify the animal near the target location. The model consists of Convolutional neural network(CNN) proving accuracy of 99.53 along with three transfer learning models of RES50,MobileNet,InceptionV3 proved to be accuracy of 99.91%.

CONCLUSION

The above surveyed data and facts indicate the decrease in number of elephants in India for several, natural and unnatural reasons, among them two important reasons ,among them the first reason is water scarcity & food shortage and whereas the second reason is the upgradation of railway tracks for high speed trains near dense forest areas. To a certain extent technological aspects , different ways with modern algorithmic methods are combining to create a concrete permanent solution to mitigate the train -elephant colloidal issue. The most important fact to enlighten is the synchronising the receiving and responding by the gadgets has to be within a very short period of time, otherwise this will led to the destruction of ecological balance. As elephants are an eco valuable species, we need to explore and implement all possible ways to protect this species from extinction.

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Saurav Bhaumik and Rakesh Kumar Mandal

Table 1. Train-Elephant Collisions in a Wide Aspect

Area	Elephant Death		Last few years Elephant Death	
	Year	Number of Elephant died	Year	Number of Elephant died
Uttarakh	1987-2001	18	2015-2020	24
Assam	1987-2006	35	2015-2019	37
Odisha	1987-2006	16	2016-2019	9
Tamil Nadu	2002-2013	13	2016-2019	2
West Bengal	1974-2015	82	2016-2019	11

Table 1. Elephant train accident cases

Railway Division	Area covered	Numberof Elephant Died
Northeast Frontier	Tinsukia, Lumding, Rangiya, Alipurduar& Katihar	25
South Eastern	Kharagpur-Adra-Ran chi-Chakradharpur	13
Southern	Chennai , Salem Division, Palakkad Division Thiruvananthapuram ,Tiruchchirapalli,Madurai Division	5
Northern	Jammu & Kashmir, Punjab, Haryana, Himachal Pradesh, Uttaranchal, Uttar Pradesh, Delhi and the Union Territory of Chandigarh,	4
North Eastern	Uttar Pradesh, Uttarakhand & western districts of Bihar	4
South Western	Bangalore & Mysore	4
East Coast	Orissa ,Some of the regions of Srikakulam, some of the districts of North Eastern Andhra Pradesh (Vizianagaram & Visakhapatnam), and some of the district of Chhattisgarh State (Bastar& Dantewada)	1
Southeast central	Bilaspur, Nagpur and Raipur	25

RESEARCH ARTICLE

Prediction of Stock Market Turbulence

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ABSTRACT

The stock market, also known as the share market, is a collection of buyers and sellers who purchase and sell stocks (also known as shares), which reflect ownership claims on companies. Stock market turbulence refers to the stock market's sudden rise and collapse. Market turbulence is frequently characterized by large movements in the stock market, both up and down. In our proposed methodology we compare two models. The first model creates a system that can detect and predict market turmoil in which the ARCH model was used to predict crisis and non-crisis occurrences with fluctuating exchange modes that indicate financial instability. Then, using the two-peak approach, it categorizes stock market crises. Then, within the framework of extended long-short-term memory (LSTM) Network, a hybrid set of standards is constructed to provide daily predictions that alert to turbulence. The second model includes GARCH (Generalized Auto Regressive Conditional Heteroskedasticity), which is used to screen the probability of the high volatility regime, and SVM (Support Vector Machine), which is used to discover signs of uncertainty risk on stock data where there is a drastic increase in volatility. Our dataset includes Indian stock market data for 30 firms from January 1st, 2000 December 31st, 2020. With a test set accuracy of 98.2% and an aggregate of two days of turbulence warning time, the first model shows promising results.

Keywords: ARCH, LSTM, Two-Peak, GARCH, SVM

INTRODUCTION

The purpose of the stock economic crisis is to predict crises by the examination of pre-turmoil patterns, allowing shareholders to take early action to minimize risks. Indeed, the scope of early warning includes the whole economic system, not only specialized global markets like banking, monetary system, and stock exchange. As a result, crisis modeling is frequently framed as a problem of categorization based on identified crisis indicators. Two aspects must

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be delicately handled in order to establish a reliable and effective system with accurate alerts and few false alarms: crisis identification and prediction mechanism.One of the most important aspects of a typical machine learning model,feature selection, has a considerable influence on prediction performance. Deep learning is a growing field of machine learning i.e allows the algorithm to extract high-level features from start to finish.The objective of this research is to present a model which can predict stock trend movement using historical data (whether the trend will be up or down).Two models are suggested as part of the investigation. Both the model uses supervised machine learning.

LITERATURE REVIEW

[1]presented a strategy for developing an Early Warning System (EWS) against mminent financial crises based on classification. The authors extend the EWS categorization approach in this model to create EWS for traditional-type crises. Long-term deterioration is the source of traditional financial crises. They carried out a case study on MFCI for the Korean financial industry. When compared to other classifiers, SVM is an appropriate estimator for such MFCI since it performs well.The fact that SVM works well for MFCI demonstrates that it is a good classifier. Given that MFCI does not utilize daily financial market data, which basically drives DFCI's behavior, it's amazing to see how they behave in the same way. This suggests that in real-life settings, two forms of economic crises (conventional and self-fulfilling crises) are inextricably intertwined. The authors of [2] suggest a novel application of deep learning models, such as paragraph vector and long short term memory, to financial time series forecasting (LSTM). The consumer price index, the price-earnings ratio, and numerous news reports are all used by investors to make judgments. Many automatic approaches to examine the data have been presented in the previous decade to assist them in making timely judgments. However, many of them employed either numerical or textual data for a single organization, but not both They propose a method that uses Paragraph Vector to transform newspaper articles into distributed representations and LSTM to assess the temporal implications of past events on opening prices for a variety of businesses in this study. The efficiency of the suggested strategy is demonstrated using real-world data from fifty Tokyo Stock Exchange-listed businesses. To get a continuous distributed representation of each news story, they use Paragraph Vector [11].The results of the experiments reveal that distributed representations of textual data outperform numerical-data-only and Bag-of-Words-based techniques. It was also revealed that LSTM outperformed other models when it came to capturing the time series impacts of input data, and that include businesses in the same industry helped forecast stock values.

The authors suggested a novel modularized stock market forecasting system (ModAugNet) in [3]. Overfitting Prevention Module and Prediction Module are two LSTM modules in Mod Aug Net. They used the ModAugNet framework, which comprises of two modules: an overfitting prevention LSTM module and a prediction LSTM module, to develop a unique data augmentation approach for stock market index forecasting. The suggested model's performance is assessed using two separate stock market data sets (S&P500 and Korea Composite Stock Price Index 200 (KOSPI200)). The results support the proposed model's good predicting accuracy. ModAugNet-c has a smaller test error than the comparator model (Single Net), which does not have an over fitting preventive LSTM module. The test mean squared error (MSE), mean absolute percentage error (MAPE), and mean absolute error (MAE) for the S&P500 were 54.1 percent, 35.5 percent, and 32.7 percent, respectively, of the corresponding Single Net S&P500 forecasting errors, while the same for the KOSPI200 were 48 percent, 23.9 percent, and 32.7 percent, respectively, of the corresponding Single Net KOSPI200 forecasting errors. Furthermore, they discovered that test performance is totally dependent on the prediction LSTM module after analysing the trained ModAugNet-c. This study has the advantage of being applicable to a variety of situations where artificially augmenting data is difficult, such as medical data analysis and financial time-series modeling. The authors of [4] provide a new hybrid time series forecasting approach that incorporates the EMD and CEEMDAN algorithms as well as the LSTM neural network. Financial time series are a kind of non linear and non stationary random signal that may be reduced across many intrinsic mode functions over a wide range of time scales using the original EMD and the complete ense-mble empiricalmode decomposition with adaptive noise (CEEMDAN). The LSTM prediction models are developed for a

ll characteristic series from EMD and CEEMDAN depositions to ensure the effect of previous data on the forecast outcome. By rebuilding each prediction series, the final prediction results are achieved. The suggested models' predictive efficiency is proved by a linear regression study of the major global stock market indices. In one step ahead forecasting of financial time series, the recommended models outperform single LSTM models, support vector machines (SVM), multi-layer perceptron (MLP), and other hybrid models. The authors created an Early Warning System to forecast financial catastrophes in[5]. Based on organized financial networks, they look at the possibility of contagion. They discover signs of contagion risk using network analysis and machine learning techniques on dates where correlations and centralities have increased significantly. The model provides a wealth of information to policymakers and investors on how to use the financial network as a tool to target assets based on centrality in order to optimise portfolio selection. Machine learning was shown to be 98.8% successful, resulting in extremely precise predictions.

The authors in [6]made a stock market trend prediction. For this objective, two models have been developed: One is for daily forecasting, while the other is for monthly forecasting. The models are built using supervised machine learning techniques. The daily for ecasting algorithm includes historical pricing and feelings. On a daily prediction model, accuracy of up to 70% has been achieved using supervised machine learning algorithms. The monthly prediction model evaluates two months' patterns to see if they are comparable. According to the analysis result, a month's trend has the least correlation with another month's trend. The authors of [7] developed a system that can detect a wide range of unanticipated worldwide catastrophes, such as terrorist attacks, disease outbreaks, and natural disasters, and feed the incident information into the model to forecast market behaviour. After experimenting for over 20 years in total on terrorist attack instances in three nations, they concluded that nightlight is an efficient indicator of market impact. The incident discovery and extraction modules, respectively, obtained 91.3 percent and 93.7 percent accuracy. The authors of [8] created a logistic regression model for improving the accuracy and efficacy of financial status indicators used to measure if scal tightness, financial market conditions, and global financial risk. The model enhances the objectivity of target variable selection.The 18 potential indicators used to create the financial status index are the 3 weighted average rate, national real estate prosperity index, money supply M2, declared effective exchange rate, and Shenzhen Component Index. From 2013 to 2017, thi s model validated China's finan-cial status.The findings show that the dynamic weighted financial condition index based on a time-varying parameter vector autoregressive model contains five variables: interest rate, real estate price, money supply, exchange rate, and stock price, all of which effectively reflect China's current financial situation. It also demonstrates that changes in financial indicators can be used to predict the degree of fiscal tightness and financial market conditions. To summarise, it is critical to watch price fluctuations, property prices, and stock prices while monitoring sy-stemic financial concerns in China. China should create an information system as well as a flexible financial regulatory supervision system to better early warning and effectively han-dle financial risks. This study is necessary for forecasting and tracking China's systemic financial vulnerabilities.

The authors suggest an early warning system(EWS) in [9], with the goal of delivering awarning signal against the possibility of mass GII sales on the local market. Theyaccomplish this by introducing a machine learning method that predicts GII behavior by forecasting future situations. Technically, this EWS is a more advanced version of Oh et alEWS. .'si[K. J. Oh, T. Y. Kim, and C.Kim] (2006). It is accomplished through the use of financial market volatility. The focusof this research is on the Korean stock market. The EWSGII project is being built in two stages. The oracle classifier is developed in the first phase, followed by the trained classifier. The second part is the creation of the lag l classifier (EWSGII). The authors of [10]try to predict and forecast prices using machine learning techniques on the cryptocurrency index and types. The aim of this article is to apply machine learning algorithms and models to predict and forecast the closing price of the cryptocurrency index 30 plus nine features of cryptocurrenc-ies, allowing users to trade these currencies more easily. They have been using a variety of machine learning techniques and method-ologies, comparing the models to determine which one gave the best results. Because of the existence of stochastic distinct elements and significant volatility, the K-NN model has not fared well in contrast to other models. The ensemble learning approach, which is the superior of all the models used here study, delivered a 92.4 percent accuracy rate.

METHODOLOGY

DATA
The data that has been used in this study is collected from nifty50 to reflect on Indian stock market turbulence. Explanatory variables used to predict stock prices are Date, Symbol, Open, High, Low, Previous close, vwrap,close,52W high, 52 week Low, Volume, Value, Number of traders. The samples span from January 1, 2000 to December 31, 2020. Fig. 1 displays the explanatory variable correlation heatmap As seen in Fig. 1, the link between most of variables is weak, contributing to the comprehensive orthogonality of records inputted. There are a few unique situations of substantial correlation that occur solely between a few variables.

The two proposed methods for analysing the data are discussed below. Method 1- First model combines the ARCH model to identify crisis/non-crisis events using high/low volatility regimes, the two-peak approach to categorize stock market crises with thresholds, and the short-time period memory (LSTM) Network to create each day's predictions. Predictions that indicate danger. volatility assumptions with conditional volatility assumptions. ARCH obviously refers to the observable fact that volatility in monetary markets is not constant—all financial records, whether or not inventory market values, oil costs, alternative costs, or GDP, experience periods of high and coffee volatility.

$$\sigma_t^2 = \alpha_0 + \alpha_1 \epsilon_{t-1}^2 + \cdots + \alpha_q \epsilon_{t-q}^2 = \alpha_0 + \sum_{i=1}^{q} \alpha_i \epsilon_{t-i}^2$$

Two-Peak Method
The two- peak technique is being developed with the end goal of determining best threshold inside binar-y classification. The lowest value between two peaks of the frequency density histogram is the binary system's optima threshold, according to the 2-peak approach. Peak was chosen for this study because it is the simplest of all and, in the end, acts as the foundation for succeeding processes. The two-peaks approach is used to establish the crisis cutoff based just on ARCH filtering probabilities of the high-volatility nation P(st = 2|Yt; t) because our crisis classifier has two states, crisis (1) and non-crisis (0). To be more exact, from time zero to time t, we define the histogram of high volatility filtering probabilities. The valley backside between theitwoifrequency tops is selected as an appropriate cutoff point at t at that factor. P(st = 2|Yt; t) c 0 if crisis = 1, otherwise. where t is the estimated parameter vector and c is crisis threshold/cutoff point.

LSTM
Long short term memory networks (LSTMs) are a type of RNN that is capable of Long shortterm memory networks (LSTMs) are a type of RNN that is capable of handling long term dependencies. Long-term dependency is something that LSTMs are designed to avoid. They don't struggle to remember data for lengthy periods of time; it's like second nature to them. volatility assumptions with conditional volatility assumptions. ARCH obviously refers to the observable fact that volatility in monetary markets is not constant—all financial records, whether or not inventory market values, oil costs, alternative costs, or GDP, experience periods of high and coffee volatility.

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Garch Model

The generalised autoregressive conditional heteroskedasticity (GARCH) process is an econometric term coined in 1982 by economist Robert F. Engle, who won the Nobel Memorial Prize in Economics in 2003. GARCH is a method for estimating financial market volatility. When attempting to predict the prices and rates of financial instruments, professionals frequently favour the GARCH process since it provides a more realistic backdrop than other models. In a statistical model, heteroskedasticity refers to the irregular pattern of variation of an error term, or variable. Essentially, heteroskedasticity exists anywhere there is volatility.The data does not follow a linear pattern. Rather, they tend to congregate. The GARCH model is a statistical model that may be used to examine a variety of financial data, such as macroeconomic data. There are three steps to creating a GARCH model in general. The first stage is to find the best-fitting autoregressive model; the second is to compute the error term's autocorrelations; and the third is to test for significance. A variety of expansions and modifications have been developed since the development of GARCH models. These variants fit neatly into one of the two types of GARCH models: symmetric and asymmetric . Let us assume some notations to express the general GARCH framework before we discuss these additions. Let t be a real-valued discrete time stochastic process, and t denotes the information set of all data collected over time t. As shown below, we define a model that includes both conditional mean and conditional variance characteristics.

$$r_t = E(r_t/\psi_{t-1}) + \varepsilon_t; \quad \varepsilon_t \sim N(0, \sigma^2_t). \quad (1)$$

Equation (1) can be rewritten as

$$r_t = \mu_t + \varepsilon_t. \quad (2)$$

The phrase $t = E(r_t/t_1)$ is used to describe the conditional mean of r_t when the information through time t is less than one. Because the error is expected to be a non-constant number over time, it is given by

εt=σtat (3)

where σt=√ V(rt/ψt−1) and at ~ N(0,1)

Keeping Equation (2) as the model for modelling the conditional mean of return, we briefly provide a number of GARCH model specifications to reflect the circumstances for expressing the conditional variance.

SVM

SVMs are supervised learning models that examine data for classification and regression analysis using integrated learning techniques. The SVM training algorithm generates a model that provides new examples in one of the two categories given a series of training examples that are individually labelled as one of the two categories. The SVM model is a set of training examples, such as space points, created in such a way that the points of the various categories are separated by a large distance. New data points are then created in same space and assigned to a category based on which side of the divide they fall into. The leading hyperplane that separates all data points from one class to the next is found via SVM. those who belong to a different category The term "leading SVM hyperplane" refers to a hyperplane with a considerable margin between the two phases . A huge slab width coupled with a hyperplane with no interior data points is referred to as a margin. The data points are supported by vectors that are close to the dividing hyperplane; these points are on the slab's edge. This could be the simplest reason for why the SVM quadratic algorithm is the best accurate machine learning method in our dataset.

RESULTS AND DISCUSSIONS

In this part, a detailed examination is carried out. The data was divided into test and training data. The training set comprises 70% of the data testing set 30% of the data. By using this evaluation metric we got an accuracy of 98.2% for the first model which incorporates LSTM and 70.6% accuracy for SVM. In the first model, after implementation of ARCH model we got volatility values between 0 and 1 , then the threshold was found using the two-peak method , that threshold was used to classify the results from ARCH model into crisis/non-crisis.The threshold is found using the histograms, already explained above. Then the results are then put as target variable with other explanatory variables in LSTM. Now, after its implementation, the model accuracy was 98.2% . In the second model, the volatility valuess from GARCH model were put into the SVM model to detect the crisis/non-crisis, the model accuracy was 70.6%. A confusion matrix was also used for SVM model accuracy. The percentages of TPR and FPR, respectively, are the proportion of correctly anticipated crisis signals over the total number of actual crises (TPR) and the percentage of incorrectly forecast crisis signals over the total number of actual tranquilly (FPR) .

$FPR = FP / FP + TN$,
$TPR = TP / TP + FN$.

The rand accuracy is the percentage of true results over the total number of instances in the assessment matrix.

$Accuracy = TP + TN + FP + FN$.
The FPR i.e, false crisis was high i.e 25 out of hundred and true positive i.e, TPR was 5 out of hundred.

CONCLUSION

Early warning systems for monetary area crises have been actively explored due to their practical relevance in providing supplemental data that supports market contributors' decision-making process as well as indicative

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information that aids policy-makers' evaluation of market susceptibility. As a result, persistent work has been made to improve the model's accuracy in terms of admonishing power and deleting bogus cautions. Two Early Warning Systems have been created, one with a novel design that integrates the ARCH model, two-peak thresholding, and LSTM, and the other with a traditional design. The GARCH model and SVM are used in the second to forecast financial exchange disruption. LSTM maintains its potency in the day-to-day forecasting function of stock issues as one of the most powerful models for dealing with time series data. To be clear, the LSTM's dependability in this study is demonstrated not only by its high accuracy of 98.2% and an average of 2 days of prior time, but also by its fundamental efficiency stability throughout the testing procedure in the test set, crossing. -follow-up and confirmation. In addition to the highly efficient prediction model, it has an accurate and reliable catastrophe identification system that contributes significantly to the EWS's efficiency and dependability.The second model, which combines GARCH and SVM, has an accuracy of 70.6 percent, making LSTM the best model for predicting stock market volatility.

In summary, we allow the LSTM-based integrated EWS to be a preferred model with two modes of operation. First, based on historical data, the EWS disaster category can be used immediately for post-date crisis detection and character categorization. The separator has been shown to work well in disaster scenarios without predicting the calamity's date or boundaries. The divider's key moment can be employed by issue models or policymakers to revisit the period of turmoil. Second, a stock crisis can be predicted using an integrated early warning system. With enough lead time and great accuracy, EWS enables market players at all levels to make quick decisions in the event of a possible emergency.

FUTURE SCOPE OF STUDY

There are numerous future directions that could be pursued. For starters, we can forecast the consequences of a wide range of events, including natural disasters and disease epidemics. Second, we can employ external knowledge bases to grasp missing features and mine news articles to generate more information. Finally, while it's difficult to capture the influence of a transitory market with current (daily) data changes, and our tests are confined to market direction estimates, we're hoping that positive market data would enable improved verification or even price guessing. Our model predicts a crisis in the next two days, with the possibility of an escalation to 4-5 days in the future.

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Table 1. evaluation metric accuracy

Methods	Procedure	Accuracy
1	ARCH +Two-peak +LSTM	98.2%
2	GARCH + SVM	70.6%
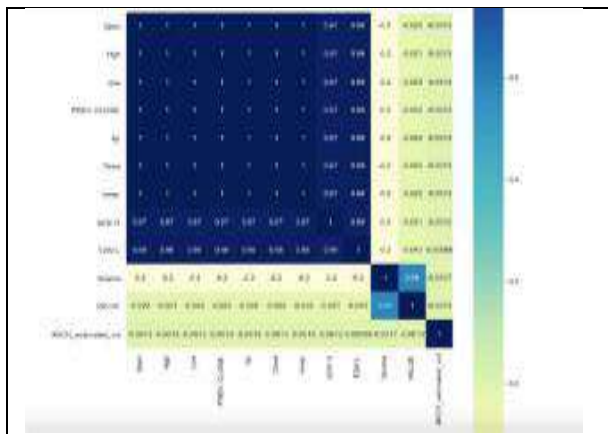
Fig 1- correlation between variables of Nifty50 data
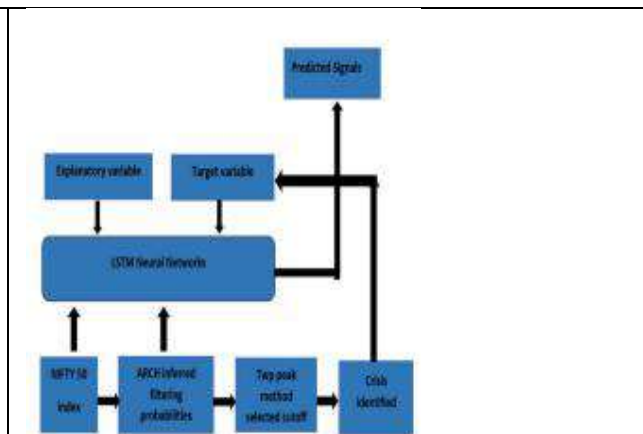
Fig2- Method 1. ARCH and the two-peak technique for identifying crises
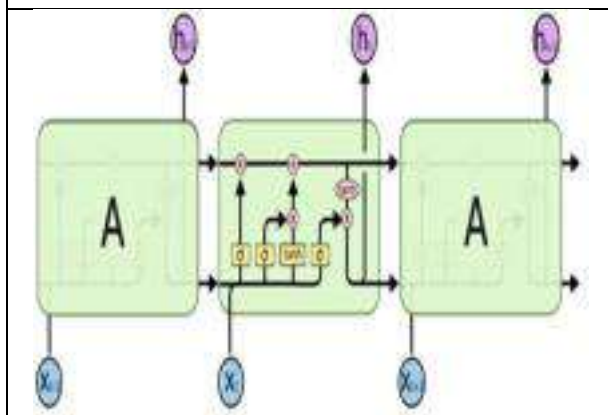
Fig 3- LSTM

Fig 4- Method 2

RESEARCH ARTICLE
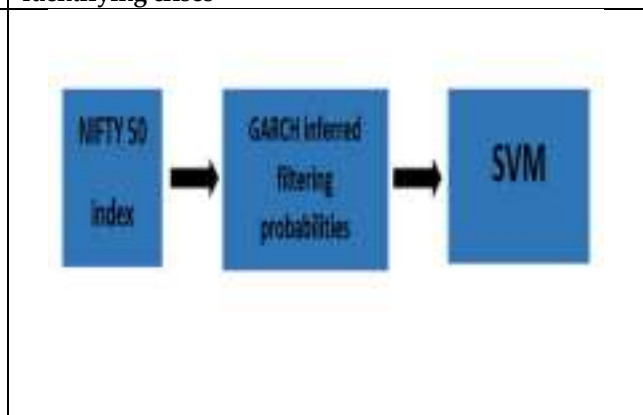
Increasing Student's Interaction and Recommendation on Learning Roadmap

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ABSTRACT

There are several challenges with e-learning, one of them is prediction on student performance and getting them to complete their subject studies. This became one of the most needful things in e-learning, so accurately predicting the student performance and classifying them based on their ongoing academic record, giving them required and necessary materials to study is critical to fulfill their on-time and satisfactory graduation so that they focus on necessary objectives. This type of problem can be solved with the help of data-driven approaches by creating a roadmap of a particular subject and providing customized learning modules to each student.

Keywords : Predicting student performance and classifying, on-time graduation, customized notes.

INTRODUCTION

The pandemic has placed a massive impact in our lives, particularly on college students' education. So for non-stop and smooth learning experience, online education is an effective answer. But this solution has quite a few drawbacks in the knowledge gaining process. We aren't aware that students will complete this assigned assignment on time and will give 100% of their effort. Since there is much less interaction between teachers, so there may be much less motivation to study and time management is absent. This isn't always the effective manner to gain knowledge and be prepared for the future. A study shows that In web-based systems, 78% of students fail to finish their courses [1]. The predominant reason of students dropping an online course is the lack of student engagement, and the second one most common reason is their incapacity to find the requisite activities and materials for the next assessment [2].
It is important to understand the problem and take necessary step to make students' life better.
The answer to these drawbacks are:
#1 Make an easy, Accessible, Interactive E-platform So that student will voluntarily access the page.

#2 Create a customized learning pattern of notes for each student.

EASY AND INTERACTIVEE-PLATFORM

A website in the E-learning platform, which acts as a significant role, because that's the sole factor that interacts with the students. Thus, if that is boring and non-interactive, students won't voluntarily access the website. And a posh website can build it arduous to scan from that website. There are few techniques which must be considered while creating an e-learning website

KIS - Keep It Simple
The online learning platform should be a straightforward and simple to load webpage. It must be accessible by any device and support any web browser.

Show Student's Analysis Report
The online platform should have necessary feature to track the student's overall performance and convey beneficial insight about their performance and study materials.

All Necessary Course Materials
The platform should not solely have notes and the written lectures. It must even have a number of videos, podcasts, recordings, Interactive tasks that students must participate, and even chats to speak with everyother.

Notification and Remainder Facility:
The platform must also have a remainder facility to warn students to complete the task within their deadline and current task to complete.

Make A Reward Based Network:
Every time a student completes his work on time, he should be rewarded with virtual-coins, so he may purchase academic merchandise with those virtual-currency. This kind of system can encourage students to complete their prep on time and gain rewards [3].

Easy Navigation
The E-platform ought to have a helping tool to navigate the website and explain all the tools accessible on the website. This will show the user how to use all the tools accurately and to its optimum potential.

CUSTOMIZE LEARNING PATTERN TO EACH STUDENT
The predominant goal of these research papers is to offer an insight about the custom designed learning pattern for every individual. In educational institution, there are diverse college students and each has their own learning pattern. If similarly type of notes is given to all of the students, then a number of them would possibly suppose it is too tough and could in no way look at the content material with inside the E-platform, however if we make it too easy, a few pupils will suppose it is a waste of time even to open the website. So In this case, students get specific and required study material in order to finish the course. Here, a topic is classified into modules which has its own weights and significance, so based on its significance we assign to the student group.
To understand this, let's take an example:

We classify learners based on some criteria and create 3 groups: A, B, C.
A group has students who barely passes the exam. B group has students who are average.
C group has students who excel the exam.
let the subject be "Operating System Concept". Here, subject can be modularized and assign weights like
Overview and history – 1.2 Basic Architecture – 3.5 Structural Components – 0.3 Scheduling Of Algorithm – 2.1
Synchronization Process – 1.1 Memory Management – 0.8 secondary storage structure- 1.0

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Each Module has its own pre-defined weight. So maximum weight signifies it is more important. For group A who are weak in learning, they will be emphasis on modules which weights more (i.e., Basic Architecture and Scheduling Algorithm) than other modules. Notes and explanation details for those modules will be thorough and clear to understand. This will make the student specialize in that particular field. Similarly, for C group who are excellent in education, for them higher level notes will be provided will more content and deeper technical knowledge. For B group and average level, notes will be provided, so that all student get only required and stress-free education. Due to customized notes, the student will voluntarily read the content provided[4]. This type of effective learning will make a lot easier to handle the course-content to the student's and give them less burden of study, which will help to increase knowledge gaining process. This knowledge will be precise and complete on a specific topic.

IMPLEMENTATIONPROCESS

For this implementation, we tend to follow seven stages of machine learning framework (1.Problem Definition, 2.Data Collection, 3.Data Preparation, 4.Visualization Of Data, 5.ML Modeling, 6.Feature Engineering, 7.Model Deployment)[5].

Problem Definition

As we mentioned earlier, the problem is lack of engagement in web-based learning. So the answer is to give only needed and customized study materials, as discussed earlier. Here, initial, we'd like to categorize the student based on their ranking. Then provide customize learning roadmap, thus we require a hybrid machine learning model. One to classify the student and therefore, the other to recommend the modules.[6]

Classification Problem

In main issue in classifying students is that we are unsure of their capability and one test cannot determine their capacity of their learning.

Recommendation System

In order to recommend best and suitable modules to the student, one must possess the domain knowledge about the topic, so that he/she can determine the appropriate weight to each module. We use content based filtering to accomplish this task [7]. For our convenience, We will only discuss the problem regarding the classification, Recommendation can be seen in further reports. The solution to this problem is quite simple, we can assess a student ability before the taking the course and after the completion of the course, so that we can understand their capability.

Data Collection

We Collected the Data from a group of college students of 'Kristu Jayanti College' who're pursuing BCA [Bachelor's in Computer Application]. They took a test on a particular subject, "Operating System Concepts & UNIX/LINUX". This was a pre-course test to perceive the simple idea of the subject from the college students. This test became primarily based totally on indistinct expertise on operating system, and the test was split into 10 questions. Each question has its very own weights based on its importance. The questions are divided into 3 levels easy, average, hard. The first four questions were of average category and weight are assigned as 0.5. The next three questions were of easy category and weight are assigned as 0.3 The last three question were of hard category and weights are assigned as 0.9. We also considered the time taken to complete the test. The total data collected was of 266 Examples and 18 Features. (266,18).

Data Preparation

Before supplying the data to the model, it must be cleansed and filter for any abnormalities. It is a crucial step in order for the model to accurately predict the result. This cleaning can be done following few steps. Find out any missing value, remove duplicate or irrelevant observations, fix structural errors, filter unwanted outliers. [8]

Missing Data

As a first option, you can drop observations that have missing values, but doing this will drop or lose information, so be mindful of this before you remove it. As an alternative option, you can supply missing values based on other observations. Again, there is an opportunity to lose the integrity of the data because you may be operating from assumptions and not actual observed data. As a different option, you would possibly adjust the manner the data is used to effectively navigate null values [3].

Below we can see how data is missing

going to remove the null data. Since it is the best way to solve this type of problem when we are dealing with credibility of student.

Fixing Irrelevant Observation

In this, using domain knowledge, find out which of the features are not required and unnecessary. Since in our case it is a primarily collected data, we collected what was required.

Data Visualization

To better understand the data, we visualize it. Since humans are visual creatures and visual images impact our thinking and improve our thought process. This eventually will help us to better understand the data and extract necessary and important feature for our model [8]. The tools used for visualization are Seaborn and Matplotlib. We generate a Heatmap to find correlation between the data. The correlation shows how data are related to each other. In this, We can see that majority of the questions are not correlated, so this says that all the questions are not related at all. This provides us obscure knowledge that the student possesses about the topic. The above plot shows us that how many students answered average level questions. From this we can signify that who scored in range 6.5 -10 has answered average level question. This plot shows that student who answered easy level questions. From this, we can signify that who scored 4-10 has answered these questions. The above plot signifies that who answered the hard level questions. From this, we can conclude those who scored 8-10 has answered a hard level question.

Feature Engineering

In this process, we have to use domain knowledge in order to search out the useful and impactful features. With the help of visualization and domain knowledge, ML engineer should use its own wit and experience to find out what features holds more weight age and what features can be removed in order to boost accuracy of model [9]. For our dataset, we are able to add the necessary weights we mentioned above. (Average level-0.5, Easy-0.3 , Hard-0.9). Those question which was answered we can assign those value and can obtain few more insight. Here we can also use time taken to complete the test. This is also an essential feature while classification of student performance. From this plot we can say that as the marks increases, the time taken to complete exam also increase. So both exam and time taken are positively correlated ($\propto$=0.56).

Label Encoding

To improve our model accuracy, we are going to encode our features. This will help machine learning model to understand the data better and feature which impact our result will be easily shown. Encoding can be done using sklearn preprocessing tools.

Normalization of Data

This means to convert data into comparable format so that all the values in the data lies between 0 -1. This will also increase accuracy of the model [10].

ML Modeling

Here, A Machine Learning Model is selected and data is supplied to the model. Selection of ML model depends on the data collected and type of result we are aiming for. In our case, we want to classify students and recommend them a tailor made course for a subject. So we can choose a classification problem combined with a recommendation

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problem. For our convenience, we will first discuss the classification problem [11]. First, we have to identify how many clusters we are going to consider. In order to do this, We will use K-Means algorithm to identify the cluster. To identify K, we are using elbow method.

As we can see that the optimal k value can be 3.
We classified the student based on their performance, and we got 3 clusters of it. Based on what we conclude in data visualization phase, as time increases, marks also increase. Based on this assumption, blue dots in graph signifies high grade students, red dots – average grade students, green dots – below average grade students.

CLASSIFICATIONALGORITHMS
In order to find the best fitting model for our data, we consider few ML algorithms[11].

Decision Tree Classifier
DTC is supervised learning method which does not have any parameter used for classification and regression. The main purpose is to make the model that predicts the value of a goal variable through learning simple decision rules inferred from the statistics features. A tree may be visible as a piece sensible constant approximation

K Nearest Neighbor
The k-nearest neighbors' method is a completely simple, easy-to-put into effect machine learning algorithm that may be used to remedy both classification and regression problems. It is a supervised based learning.

Linear Discriminant Analysis (LDA)
A classifier with a linear decision boundary, generated with the aid of using fitting class conditional densities to the facts and the usage of Bayes' rule. The model fits a Gaussian density to every class, assuming that each one classes share the same covariance matrix. The geared up model also can be used to lessen the dimensionality of the enter with the aid of using projecting it to the maximum discriminative directions, the usage of the transform method[12].

Quadratic Discriminant Analysis (QDA)
It is a classifier with a quadratic decision boundary, generated through becoming class conditional densities to the records and the use of Bayes' rule. The model fits a Gaussian density to every class[12]. Let us find how does this algorithm fit the data. The model was supplied with the train data and preformed Repeated Stratified K-Fold with n=2, so that the model learns the data.

As we can see from above, the Decision tree got an accuracy of 86.7% and least of 75.75%.
K Nearest Neighbor
K Nearest algorithm got the highest accuracy of 82.9% and least is 76.89%[12].

LDA Algorithm
LDA Algorithm got the highest accuracy of 91.2% and least of 85.98%[11].
QDA Algorithm
QDA Algorithm got the highest accuracy of 90.53% and least of 83.33%[12].
Let us compare classifier with actual predicted value.

KNN algorithm's accuracy has a lot of deviation, so this is less suitable for this type of problem. Classification Tree is shows significant growth when sample size is less, But as sample increase the accuracy decrease [11]. LDA Algorithm also shows significant increase of growth and deviation in accuracy is also less. QDA Algorithm Show upward and smooth movement in accuracy. Both LDA and QDA are Ideal for this type of multiple classification of data.

ROC and Precision Call

The LDA Algorithm has much consistent and accuracy is pretty higher than other algorithms[11]. The above can be summarized in a tabular format

CONCLUSION

This research can provide beneficial impact on student behavior and their learning pattern. So eventually this enables us to provide a better roadmap to the student to study the course effectively. The ML model use in this will assist to classify the students based on their check result and deliver them higher experience in learning. Student will finally voluntarily take Initiative to learn and enhance themselves. Having an easy and Interactive UI may also boom the chance of learning in e-based learning. The reward system for completion of their work can also increase the productivity.

FUTURE ENHANCEMENT

This research paper's handiest talks about the initiative phase of creating student engage with online learning, we will cross plenty greater in element about the sub phase of the interaction. The Recommendation System for selection of course module can also be implemented. With the assist of classifying maximum weighted module first to the least weights.

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Table-1 : Highest Accuracy of A Classifier Performing of Different Accuracy Standards

Classifiers	DTC	KNN	LDA	QDA
2-Fold K	86.7%	82.9%	**91.2%**	90.5%
Cross Validation	85.9%	90.1%	**92.2%**	89.3%
Accuracy + ROC	91.3%	94.8%	**98.2%**	89.2%

Table-2: Accuracy of Classifier on Different Sample Folds.

N-Folds	2	7	12	20
DTC	89.8%	89.0%	**90.0%**	80.2%
KNN	81.5%	80.0%	**82.5%**	80.3%
LDA	89.8%	89.5%	90.1%	**92.2%**
QDA	89.2%	87.5%	89.5%	**90.3%**
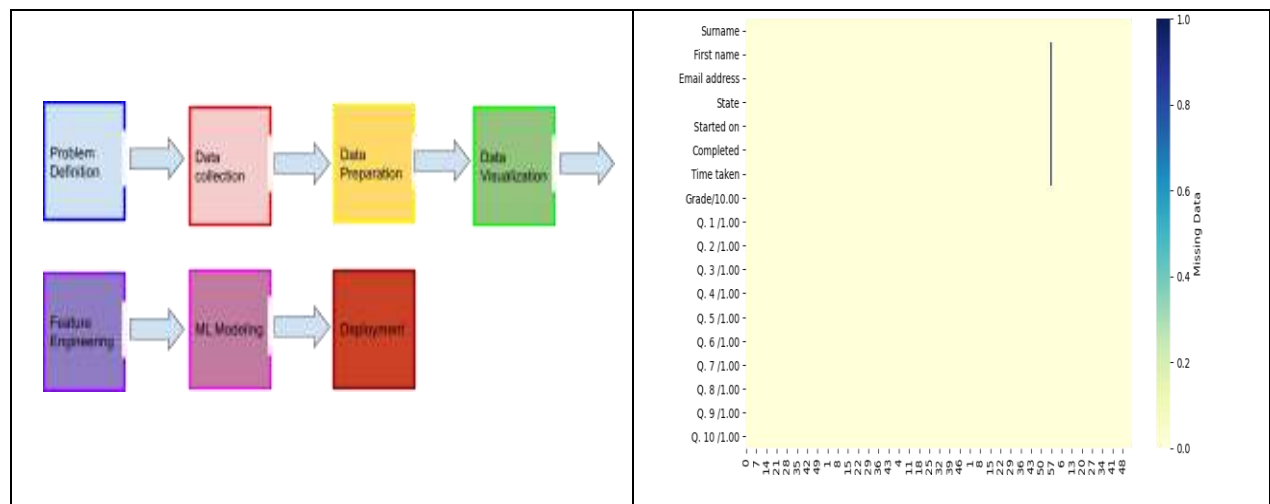
Figure 1: seven stage model	**Figure 2. Missing Data**
Figure 3: Heatmap for data correlation	**Figure 4: Scatter plot of data correlation**
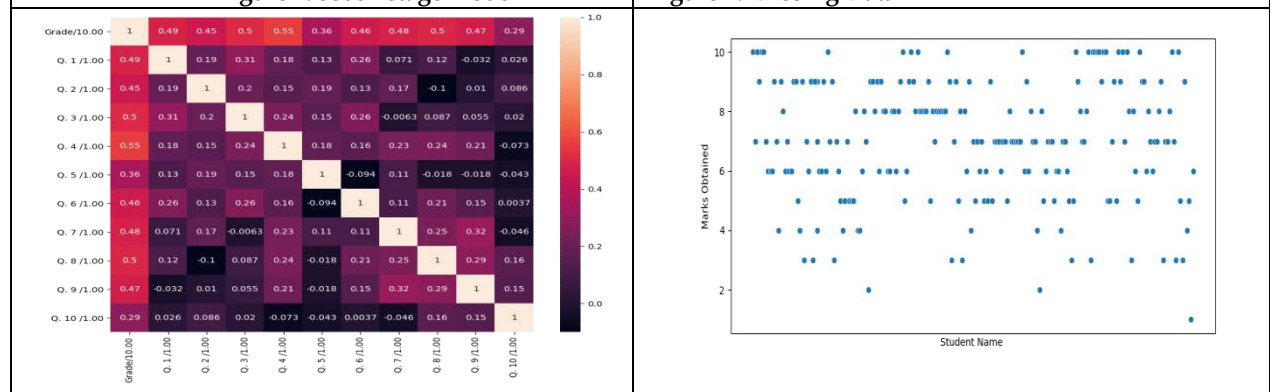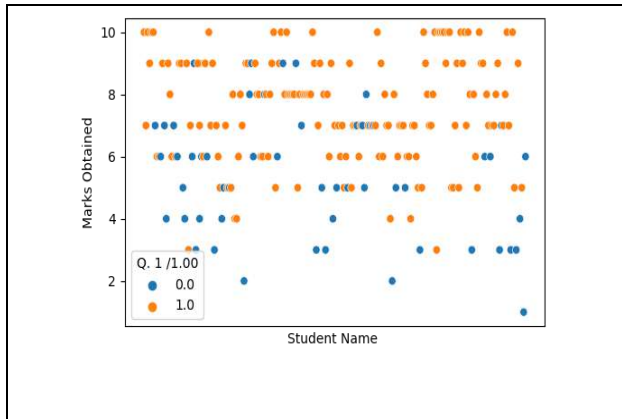
Figure 5: scatter plot of data who answered average question
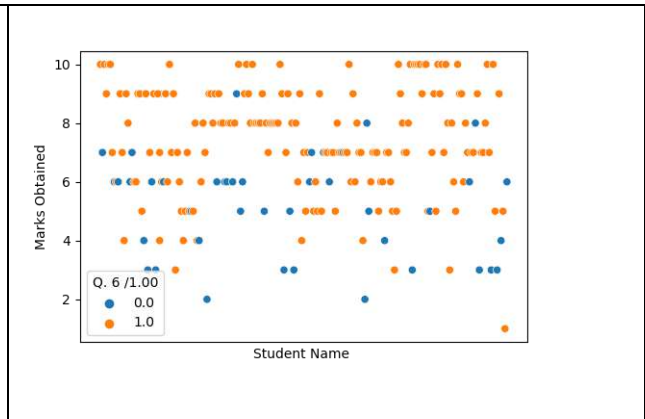
Figure 6: Scatter plot student answered easy question
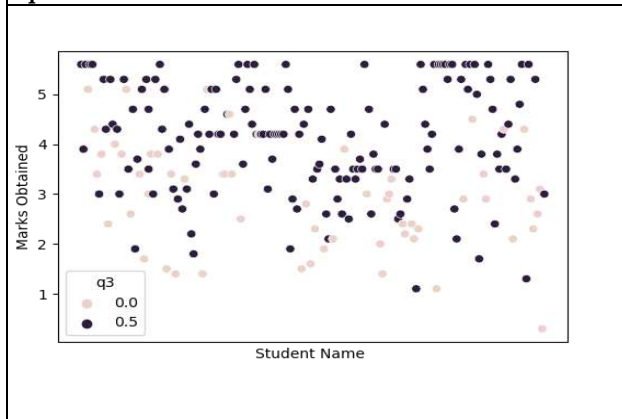
Figure 7 : Scatter plot After Adding Weights to average level question
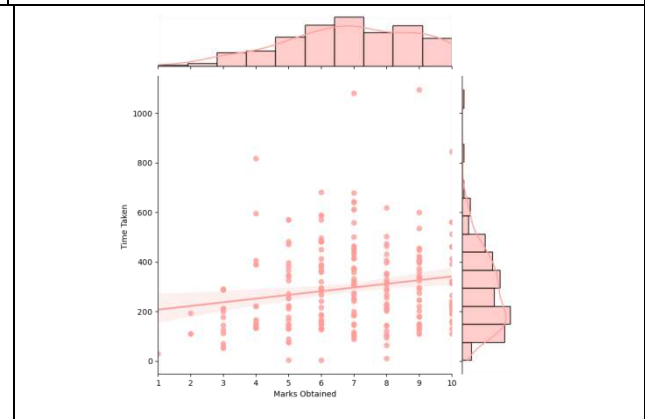
Figure 8: Joint plot on marks and time taken From
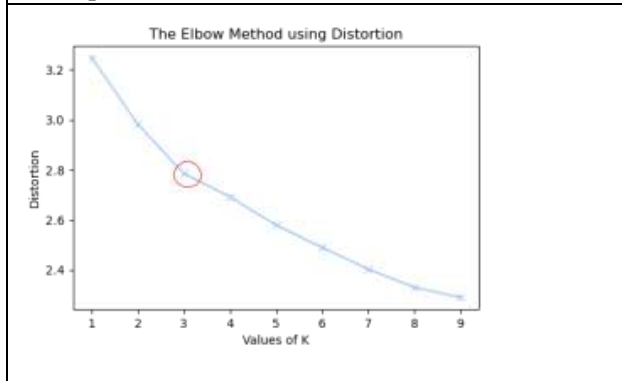
Figure 9: Elbow Method using Distortion
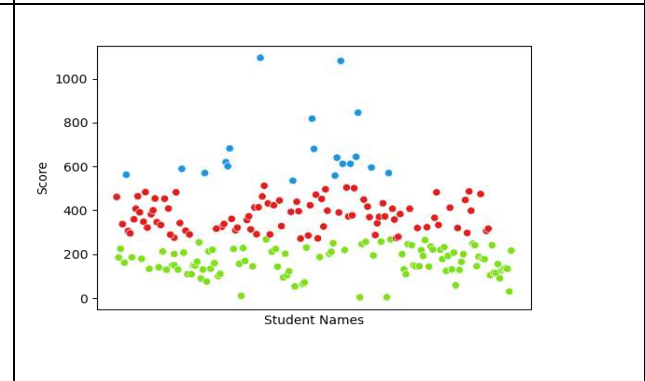
Figure 10: K-means Cluster

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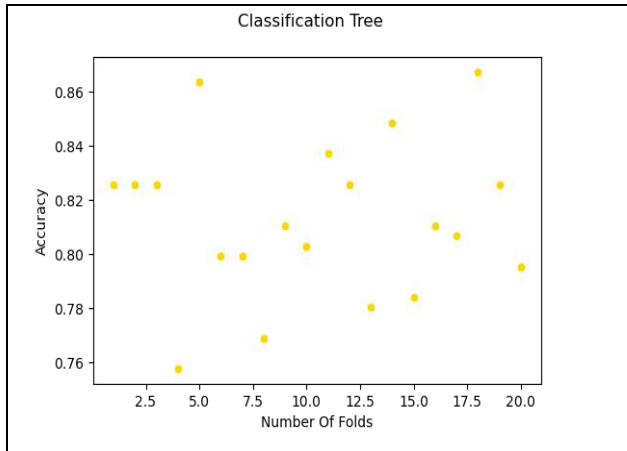
Figure 11. DTC Algorithm
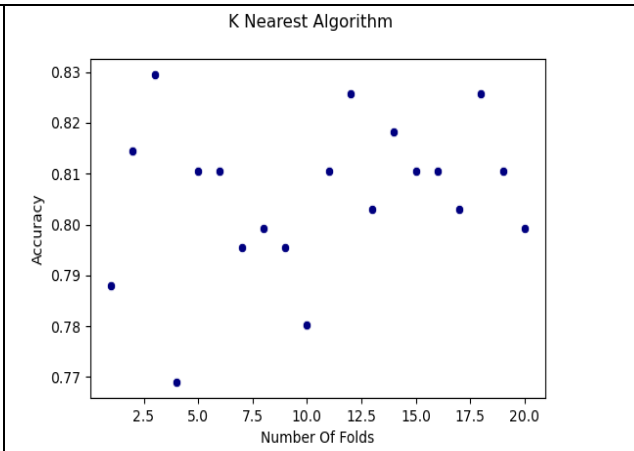
Figure 12. K Nearest Neighbor
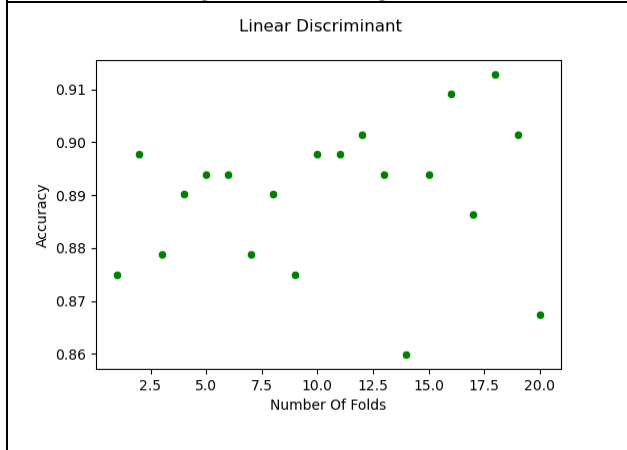
Figure 13. LDA Algorithm
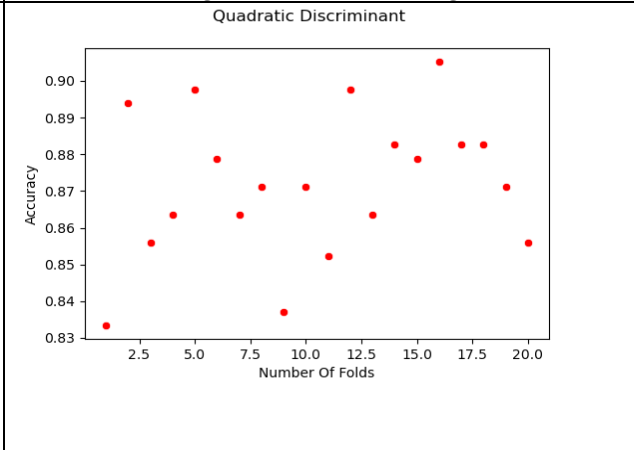
Figure 14. QDA Algorithm
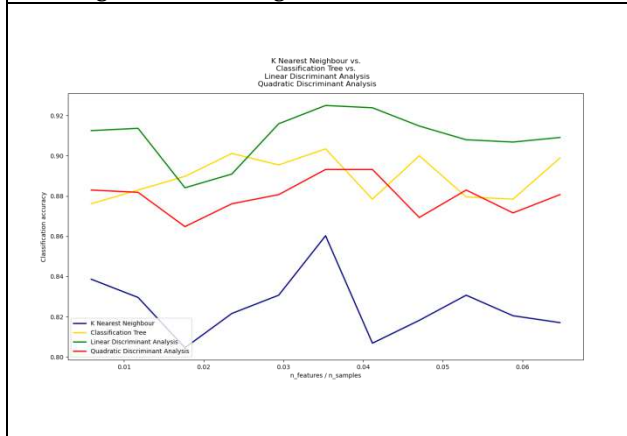
Figure 15. QDA Algorithm got the highest accuracy
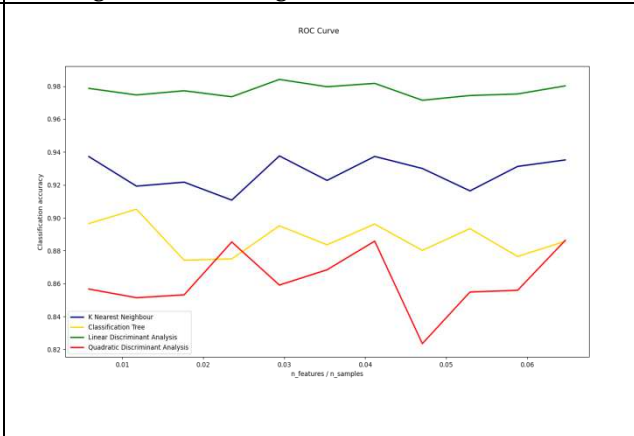
Figure 16. ROC and Precision Call

RESEARCH ARTICLE

Hypoglycemic and Hypolipidemic Effect of Hydroethanolic Extract of *Rubia Cordifolia* Linn Root Extract in Alloxan Induced Diabetic Rats

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ABSTRACT

Diabetes mellitus is a major health problem of today's world. The number of people suffering from diabetus is increasing. There are multiple therapies available to treat diabeties, but total recovery may not be possible. The allopathic drugs have adverse side effects including haematological, cutaneous and gastrointestinal reactions, hyoglycemic coma. The antidiabetic medicines from plants have a simillar activity and negligible side effect with low cost. Herbal drugs and phytoconstituent are useful for the management of diabetes. The present study is aimed to evaluate the antidiabetic and hypolipidemic activity of 50% hydroethanolic extract of *Rubia cordifolia Linn.* in alloxan induced diabetic rats. Alloxan monohydrate was used to induce diabetes in wistar albino rats (120 – 150 g). The preliminary phytochemical screening revealed the presence of various phytoconstituents. The acute toxicity study reported that LD_{50} value of root sample was 1.4 g/kg b.w. There is no change in body weight during the treatment period. Serum glucose, insulin, liver glycogen, GHb, TC, TG, LDL-C and VLDL-C levels were increased in diabetic rats. These results were reverted in plant extract treated diabetic animals. The reduced HDL level was improved significantly in extract treated groups. These results were compared with standard drug glibenclamide treated animals. The present study suggests that the plant extract had significant antidiabetic, hypolipidemic activity in alloxan induced diabetic rats.

Keywords: *Rubia cordifolia Linn.*, Alloxan, Glibenclamide, Phytochemicals, Acute toxicity study, Serum glucose, Total Cholesterol.

INTRODUCTION

Diabetes mellitus is one of the most common endocrine disorder characterized by abnormal carbohydrate metabolism that result in high blood sugar with altered lipid and protein metabolism. It occurs mainly due to defect in pancreatic insulin secretion with or without concurrent impairment of insulin action. Hyperglycemia and Hyerlipidemia are two important characteristics of diabetes in which patients experience various vascular complications such as atherosclerosis, coronary heart diseases, diabetic nephropathy, neuropathy [1]. Diabetus mellitus is associated with profound alteration in plasma lipid and lipid profile [2]. To attain strict glucose control when managing diabetes had utilized daily injection of insulin and oral hypoglycemic drugs sulfonylurea and metformin. The usage of oral drug is limited due to adverse side effects including haematological, cutaneous and gastrointestinal reactions, hypoglycemic coma and disturbances of liver and kidney function [3]. Medicinal plants are widely used for the treatment of human diseases all over the world. More than 800 plant species having hypoglycemic activity have been available in literature [4]. They contain glycosides, alkaloids, terpenoids, flavonoids and carotein etc., that are implicated as having antidiabetic effects. Herbal drugs have lesser or no side effects and are less expensive as compared to synthetic drugs. These drugs are single plant extract or fractions or mixtures of extracts from different plants which are standardized for their safety and efficacy [5]. Therefore plant materials are continuously scrutinized and explored for their effect as hypoglycemic agents

Rubia cordifolia Linn. commonly called as Manjistha or Indian Madder is a prickly creeper or climber. It is distributed throughout the lower hills of Indian Himalayas in North and Western Ghats, Japan, Ceylon, Malay and in most tropical forests up to an altitude of 3500 meters. About 15 species are found in India [6]. Important one of these are Indian Madder [7,8]. This plant is known as best blood purifying herb in Ayurveda [9]. It also has anticancer [10], astringent, deobstruent and diuretic properties. It is used to cure rheumatism, ulcers, inflammation, skin problems and leprosy. Major constituents are anthroquinone, hexapeptide, alizarin, glycosides and terpenes [11]. In the present study efforts have been made to establish the scientific validity of the antihyperglycemic and hyolipidemic property of the *Rubia cordifolia* Linn.

MATERIALS AND METHODS

Plant Collection and Authentication
The dried roots of *Rubia cordifolia* Linn. Were purchased from a well known herbal materials supplier of Tirupur, Tamil Nadu. A Voucher Specimen of the plant was collected from the lower hills area of Ooty and was authenticated by Dr. S. RAJAN, Department of AYUSH, Ministry of Health and Family Welfare, Government of India, Nilgiri District.

Phytochemical Screening
The dried powdered plant material was subjected to soxhelet extraction with different solvents such as ethanol, acetone, chloroform, distilled water, petroleum ether and benzene for 18 hours. The extracts were condensed by rotary evaporator and were used for screening of phytochemicals.

Plant extract preparation
About 5 kg of plant root was pulverized using a mixer grinder. The coarse powder was used for the preparation of the extract. The coarse powder was cold macerated with ethanol (i.e. 2 litres of ethanol and 2 liters of water) for 3 days, with occasional stirring. After 3 days the suspension was filtered through a fine muslin cloth and the residue was removed. Then the water portion of the samples was evaporated at a low temperature (approximately at 40^0C) under reduced pressure in a rotary evaporator. Dark brown coloured crystals of 0.9 gm was obtained. The crystals were stored in an air-tight dessicator and whenever needed, the residues were dissolved in distilled water, filtered (whatman No.1 filter paper) and used for the studies.

Experimental animals

Male albino rats of Wistar strain weighing 120 – 150 g were procured from the animal house, PSG Institute of Medical Science and Research (No: 158/1999/CPCSEA), Coimbatore, India. The rats were grouped and housed in polyacrylic cages and maintained under standard conditions (25 ± 2^{0}C) with 12 ± 1 hours dark/light cycle. The animals were fed with rat pellet feed supplied by Hindustan Lever Ltd., Bangalore, India and water *ad libitum.* All the experimental procedures were conducted after the approval of ethical committee and were in strict accordance with institutional animal ethical committee guidelines for the care and use of laboratory animals.

Induction of diabetes

Alloxan monohydrate was used to induce diabetes mellitus in normoglycemic rats. Animals were allowed to fast for 18 hours and were injected intraperitonially with freshly prepared alloxan monohydrate in sterile normal saline in a dose of 120 mg/kg body weight. After 72 hours of injection, 0.5 to 1 ml of blood was collected from the rats using microcapillary tube by retro-orbital route. The diabetic rats with fasting blood glucose levels greater than 350 mg/dl approximately were selected for the study.

Acute Toxicity Study

Wistar albino rats were fasted overnight, weighed and divided into 6 groups, each group consisting of five animals. Test samples were administered separately in various doses by oral route (2000, 4000, 6000, 8000, 10000 mg/kg body wt). One group was maintained as normal control and was given vehicle alone. After oral administration of the extract, the animals were observed continuously for the first two hours for death due to acute toxicity. The results of LD$_{50}$ study were analyzed using Miller and Tainter [12] method.

Diabetic Treatment groups

The experimental rats were divided into 6 groups of 6 animals in each group, after two weeks of acclimatization period.

Normal Control	-	Healthy untreated rats.
Diabetic control	-	Diabetic rats.
Glibenclamide Control	-	Drug Control (glibenclamide 600 µg/kg body weight)
Treatment I	-	Diabetic rats received 50% hydroethanolic extract of *Rubia cordifolia* Linn. (root) extract (400 mg/kg body weight) for 28 days.

Blood sample collection

At the end of treatment period (28 days), blood samples (1 – 1.5 ml) were collected after mild chloroform anesthesia and by the cardiac puncture. Serum was collected from blood by centrifuging at 3000 rpm for 20 minutes at room temperature (for enzyme assays blood was centrifuged at 40C). Body weight of rats were taken before and after treatment.

Biochemical investigation

The condensed extract were used for screening phytochemicals such as alkaloids (Iodine, Wagner and Dragendroff's test), flavonoids (pew's, Shineda and NaoH test), glycosides(Keller – Killiani, conH2SO4) and Molisch test), phenols(FeCl3, Leadacetate, Libermann's test), Steroids(Libermann- Burchard's, Salkowski test), Tannins (FeCl3, Lead acetate test), Total carbohydrate(Fehling's, Benedict's test), Total Protein(Biuret, Million's test), Saponins(foam, Haemolysis test).Serum glucose was estimated by glucose oxidase (GOD) peroxidase (POD) kit method [13]. Estimation of liver glycogen [14] , Insulin [15], Glycosylated Hb [16,17], Total cholesterol [18], HDL cholesterol [19], Triglycerides [20]. The LDL, VLDL values were calculated by Friedewalds formula [21] VLDL = TG/5, LDL = TC-(HDL+VLDL).

Statistical analysis

Data were reported as means ± SD by using the statistical package of social sciences (SPSS) version 10.0 for windows. Data) were analyzed using analysis of Variance (ANOVA) and the group means were compared by Duncan's

Multiple Range Test (DMRT). Values were considered statistically significant when P < 0.05 and thus the groups were compared as given in the table.

RESULT AND DISCUSSION

PHYTOCHEMICAL SCREENING OF *Rubia cordifolia* Linn. root.
The preliminary phytochemical screening of plant extracts were shown in table 1. The root extract of *Rubia cordifolia* Linn. revealed the presence of alkaloids, flavonoids, glycosides, phenols, saponins, tannins, carbohydrates and protein and showed absence of safronins. These phytochemicals has been reported to exhibit multiple biological effects including anti-inflammatory, antitumor activities [22]. From the preliminary phytochemical screening tests, it was clear that water and ethanol extract of plant sample showed presence of biologically important phytoconstituents. This observation may be due to two reasons firstly, the nature of bio-active components could be enhanced in presence of water and ethanol. Secondly the stronger extraction capacity of water and ethanol could have produced greater amount of active constituents. The importance of medicinal plants were considered in terms of their phytoconstituents. Thus the preliminary screening tests are useful in the detection of bioactive principles and may lead to discovery and development of new drugs

Acute Toxicity Study of 50% Hydroethanolic Root Extract of *Rubia cordifolia* Linn.
The toxicity determinations were done for the doses of 2, 4, 6, 8, 10 g/kg b.w. The results were tabulated as shown in table 2. When a plot of log dose (on X- axis) vs % mortality (on Y- axis) was plotted, a line was obtained. The log dose corresponding to probit 5 was determined and its corresponding antilog value was computed which is taken as the LD_{50} value of the sample. The LD_{50} value of root sample was found to be 1.4 g/kg b.w.

Effect on Body Weight
All the animals from treated groups did not show any significant decrease in body weight following 28 days of treatment as compared with 0 day treatment. The experimental animals showed slight increase in body weight which was statistically insignificant (table 3). This effect of the dose may be due to its non- nutritional value of plant sample. Body weight is an important factor to monitor the health of an animal. From the result of this study, it was observed that at all normal therapeutic doses, the test sample was considered to be safe for long term oral treatment.

Antidiabetic and Hypolipidimic Activity of Plant Extract
Effect on Blood Glucose
Table 4 describes the effect of treatment of plant extract on serum glucose level. Diabetic rats showed significant increase in serum glucose level by 38% whereas glibenclamide, *Rubia cordifolia* Linn. treated diabetic rats showed reduction in glucose of 53% and 46% respectively.

Effect on Glycosylated Hb.
Table 5 shows treatment with glibenclamide and *Rubia cordifolia* Linn. plant extract reduced percentage GHb levels from 19.8% (DC) to 10.5% and 12.3% respectively. Serum Glycosylated Hb level represents average value of blood glucose over last or next 3 to 4 month period. This level provides useful means of assessing treatment efficacy [23]. The results indicate that plant extract prevented the elevation of glycosylated Hb in diabetic rats. This could be due to better glycemic control produced by the plant extract. Similar result has been seen with the plant *Sphaeranthus indices* Linn [24]**.** extract in diabetes induced rats.

Liver Glycogen and Insulin
Diabetic rats had 4.2 times lowered glycogen content (76% lowered) when compared to normal control rats (table 6**)**. The treatment group showed (120%) improvement in glycogen level significantly (P<0.05). The decrease in liver glycogen content in diabetic state might be due to the lack of insulin which leads to inactivation of glycogen synthetase system. The diabetic control rat showed 48% reduction in insulin level compared to normal control rats.

Treatment with plant extract showed increase in insulin level. The results indicate that plant extract treatment leads to reactivation of glycogen synthetase system by increasing insulin secretion in treated animals [25]. This results are well correlated with the findings of *Barleria prionitis* Linn [26]. and *Momordica charantia* fruit extract [23] treatment to diabetic rats.

Lipid Profile

Figure 1, 2 and tables 7, 8, 9 describe the effect of plant extract and standard drug glibenclamide on serum lipid profile. An increase in serum TC, TGs, VLDL-C and LDL-C levels whereas decrease in HDL-C level were observed in diabetic rats when compared to normal control rats. Significant reduction in total cholesterol (Figure 1) was observed in drug glibenclamide and *Rubia cordifolia* Linn. after 28 days of treatment. In *Rubia cordifolia* Linn. treated rats TG level were reduced to 45.7% and HDL- C level increased to 119.8%. Administration of plant extract decreased the elevated LDL-C level in diabetic rats when compared to diabetic untreated control rats (figure 2). Diabetes affects both glucose and lipid metabolism [27]. The insulin deficiency depletes the activity of lipoprotein lipase, thus leading to altered lipoprotein metabolism during diabetes [28]. Insulin has a potent inhibitory effect on lipolysis in adipocytes. Insulin deficiency leads to excess lipolysis and increased influx of free fatty acids to the liver [29,30]. The increased levels of LDL-C and VLDL-C in diabetic animals might be due to stimulation of hepatic triglyceride synthesis as a result of free fatty acid influx. In diabetus the HDL-C level was reduced significanty ($P<0.05$) indicating positive risk factor for atherosclerosis [31]. Results of the present study reveals that diabetic rat treated with plant extract recorded better depletion of TC, TG, LDL - C, VLDL-C and increase in HDL-C level. This might be due to reduced hepatic triglyceride synthesis and or reduced lipolysis due to increase in serum insulin levels. Improvement in HDL-C level indicates a reversed atherogenic risk in plant extract treated groups. This results are well correlated with the findings on *Morus indica* and *Asystasa gangetica* [32], *Bougainvllea glabra* [33].

CONCLUSION

Phytochemical screening proved the presence of various bioactive components in the plant extract. The extract produced significant antidiabetic, hyolipidemic activity in diabetic rats. The results are comparable with standard drug glibenclamide. The plant had high medicinal value and may be recommended as cheap food supplements in treating diabetes, hyperlipidemic conditions after a thorough development in this aspect.

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Table 1. Phytochemical screening of *Rubia cordifolia* Linn. root extract.

Phytoconstituent	Alcohol	Acetone	chloroform	Distilled Water	Petroleum ether	Benzene
Alkaloids	+ + +	+ + +	- - -	+ + +	+ + +	- - -
Flavonoids	+	+	+	+	-	-
Phenols	+	-	+	+	-	-
Tannin	+ +	+ +	- -	+ +	- -	- -
Glycosides	+ +	- -	- -	+ +	- -	- -
Saponin	+ +	+ +	- -	+ +	- -	- -
Safronins	-	-	-	-	-	-
Total carbohydrate	+ +	+ +	- -	+ +	+ +	-
Total protein	+ +	-	- -	+ +	- -	- -
Steroids	+	-	-	+	-	-
Thiols	-	-	-	+	-	-

+ , - symbols indicate the presence and absence of phytochemicals respecticely.

Table 2. Miller and Tainter method of estimation of LD_{50} for *Rubia cordifolia* Linn. root extract

Groups	Dose (g/kg b.w)	Log Dose	Dead Total	% Dead	Corrected %	Probit
Group-I	2	0.3010	0	0	5	3.36
Group-II	4	0.6020	0	0	5	3.36
Group-III	6	0.7781	0	0	5	3.36
Group-IV	8	0.9030	1	20	20	4.16
Group-V	10	1.0000	2	40	40	4.75

Table 3. Change in body weight of rats following the 50% hydroethanolic root extract

Group	*Rubia cordifolia* Linn.	
	Before Treatment	After Treatment
Control	146.67 ± 2.87	158.00 ± 9.54
Group-I	140.00 ± 8.10	151.00 ± 2.77*
Group-II	142.00 ± 4.65	153.00 ±2.95*
Group-III	148.00 ± 4.90	150.00 ± 3.01*
Group-IV	145.00 ± 10.00	155.00 ± 2.70*
Group-V	146.00 ± 0.00	157.00 ± 2.70*

P<0.05 when compared treatment to corresponding control groups.   *-statistically not significant

Table 4 Effect of plant extract on blood glucose level.

Groups	Day-0	Day-7	Day-14	Day-21	Day-28
Normal Control	98.3 ± 7.10	96 ± 6.83	98.3 ±9.5	99.67 ± 9.94	102.67 ± 6.94
Diabetic Control	266.4 ± 3.5	282.7 ± 7.0	299 ± 11	346.77 ± 7.0	366.6 ± 12.21
Glibenclamide control	253 ± 19.3[a]	192 ± 0.0[a]	168± 1.2[a]	144.4 ± 1.02[a]	118 ± 8.20[a]
Treatment I	292 ± 26.5[b]	248 ± 9.7[b]	227 ± 3[b]	210.7 ± 34.8[b]	157.7 ± 18.7[b]

Treatment I - (*Rubia codifolia* Linn. root, 400 mg/kg b.w). a- comparison between diabetic control with Glibenclamide group is significant at 5% level.   b - comparison between diabetic control with Treatment group I is significant at 5% level

Table 5. Effect of plant extract on glycosylated Haemoglobin level

Groups	Glycosylated Hb (mg%)
Normal Control	6.2
Diabetic Control	19.8
Glibenclamide Control	10.5
Treatment I	12.3

Values are mean ± SD of six samples. P< 0.05 as compared to diabetic control. Treatment I (*Rubia codifolia* Linn. root, 400 mg/kg b.w.

Table 6. Effect of plant extract on liver glycogen and insulin level.

Groups	Liver Glycogen (mg/g tissue)	Insulin (µU/ml)
Normal Control	58.09 ± 0.08	23.6 ± 4.5
Diabetic Control	13.84 ± 0.03	12.2 ± 1.3
Glibenclamide Control	40.17 ± 0.03[a]	17.9 ± 6.4[a]
Treatment I	30.84 ± 0.02[b]	14.7 ± 4.9[b]

Table 7. Effect of plant extract on the level of Triglycerides.

Groups	Triglycerides (mg/dl)				
	Day-0	Day-7	Day-14	Day-21	Day -28
Normal Control	124 ± 5.8	131 ± 7.2	125.3 ± 4.7	127 ± 5.7	126 ± 11
Diabetic Control	199.7 ± 9.4	210 ± 4.1	230 ± 2.4	243.5 ± 14.5	264.1 ± 3.1
Glibenclamide Control	196 ± 8.4[a]	181 ± 1[a]	155.9 ± 13[a]	130 ± 11.0[a]	112 ± 15.2[a]
Treatment I	188 ± 7.1[b]	176 ± 13[b]	165 ± 13[b]	156.67 ± 2[b]	143.2 ± 4.[b]

Values are mean ± SD of six samples. P< 0.05 as compared to diabetic control.

Treatment I (*Rubia codifolia* Linn. root, 400 mg/kg b.w),

a - comparison between diabetic control with Glibenclamide group is significant at 5% level

b - comparison between diabetic control with Treatment group I is significant at 5% level

Table 8. Effect of plant extract on the level of HDL-cholesterol.

Groups	HDL-Cholesterol (mg/dl)				
	Day-0	Day-7	Day-14	Day-21	Day -28
Normal Control	39.0 ± 2.41	40.7 ± 0.89	41.0 ± 1.02	40.3 ± 1.47	39.1 ± 4.05
Diabetic Control	30.0 ± 1.82	27.3 ± 3.36	25.2 ± 5.10	20.0 ± 1.89	19.1 ± 4.80
Glibenclamide Control	32.3 ± 4.36[a]	40.0 ± 3.49[a]	45.14 ±5.0[a]	48.67 ± 2.0[a]	54.6 ± 7.20[a]
Treatment I	31.0 ± 1.60[b]	35.7 ± 5.73[b]	38.3 ± 1.42[b]	40.0 ± 1.2[b]	42.0 ± 3.40[b]

Table 9. Effect of plant extract on the level of VLDL -cholesterol.

Groups	VLDL -Cholesterol (mg/dl)				
	Day-0	Day-7	Day-14	Day-21	Day -28
Normal Control	27.0 ± 0.14	28.6 ± 1.9	29.9 ± 1.85	31 ± 1.80	29.73 ± 0.2
Diabetic Control	42.4 ± 2.1	46.47 ±4.6	48.6 ± 5.3	50.53 ± 4.2	58.87 ± 2.3
Glibenclamide Control	43 ±1.87[a]	42 ± 1.09[a]	36.3 ± 1.05[a]	33.3 ±1.05[a]	27.4 ± 1.1[a]
Treatment I	44 ± 5.3[b]	43.2 ± 1.4[b]	41.9 ± 1.3[a]	38.96 ±6.2[b]	35.2 ± 2.6[b]

Values are mean ± SD of six samples. $P < 0.05$ as compared to diabetic control.

Treatment I (*Rubia codifolia* Linn. root, 400 mg/kg b.w),

a - comparison between diabetic control with Glibenclamide group is significant at 5% level

b - comparison between diabetic control with Treatment group I is significant at 5% level
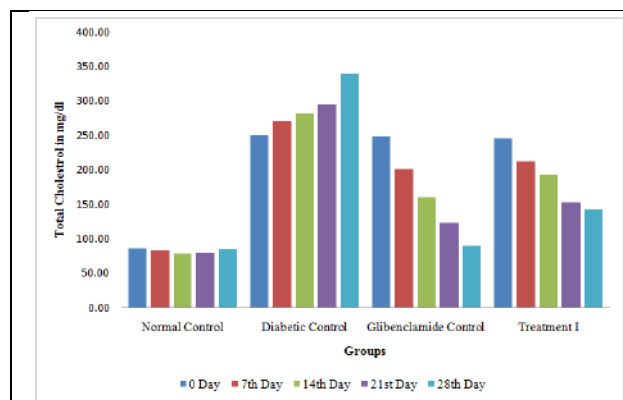
Figure 1 Effect of plant extract on the level of total cholesterol **Treatment I (*Rubia codifolia* Linn. root, 400 mg/kg b.w).**	**Figure 2 Effect of plant extract on the level of LDL -cholesterol** **Treatment I (*Rubia codifolia* Linn. root, 400 mg/kg b.w).**
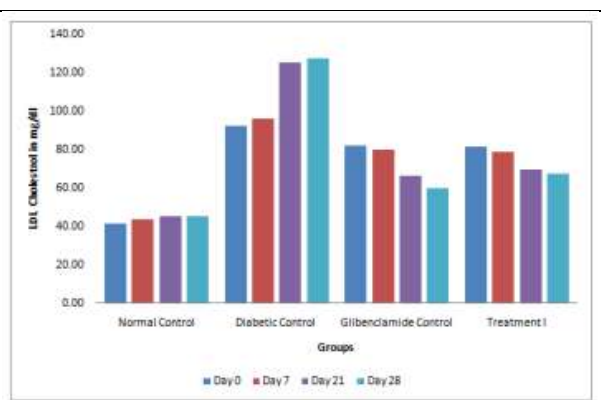
RESEARCH ARTICLE

Frequency Transients in A Solar PV Microgrid Under Varying Step Solar Radiation Levels

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ABSTRACT

In this work, PV based rural microgrid is taken to assess transients performance of the system subjected to solar irradiance variation. For examining the transients two control variables are considered which are solar irradiance level and load power factor. The system taken for the study is a AC system with phase voltage variation of 90-250 Volts. The model is simulated and analysed in MATLAB Simulink platform to understand the role of these control variables in affecting the frequency peaks of the PV microgrid.

Keywords: Solar PV Microgrid, Frequency transients, Simulink model analysis, Variable Control parameters.

INTRODUCTION

Any variation in power frequency is defined as the variation that has coccurs with respect to fundamental frequency that can be 50 Hz or 60 Hz based upon the standard of operation. In traditional grid operated systems with conventional power sources such deviations result due to sudden causes like faults in the line, connection or disconnection that place suddenly encompassing large value of loads, or any generators that are connected in the system becomes unplanned offline. Now a days frequency variation is a rear incidence that may come in conventioanal sourced based power plants however, in the case of solar PV or other renewable energy based distribution generation, the use of inverters and MPPT may affect the frequency transient performance under various types of input disturbances, particularly in view of the fast switching operations of power electronics converters used in both low and medium voltage systems. When the analysis of frequency transients is to be studied, a suitable model is required to give a proper response estimation of the high frequency transients generated mainly due to the combined effects of the variation of control inputs and switching operation of the power electronics

converters used. *Matlab-Simulink* models have been widely used for transient studies in power systems [1]-[6]. Thus, as the *Matlab –Simulink* model has been shown to be versatile for power system transient studies, we feel that it can be suitable for modeling frequency transients in a solar PV microgrid.

The Solar PV Microgrid System

A general architecture of solar PV system will integrates PV array with an MPPT to track the operating point. Along with it DC-DC converter is added to regulate PWM signal for MPPT and then it connectes to inverter to supply the electricity to the connected AC loads. Figure 1 shows a simplified schematic of the solar PV microgrid proposed for the study. Figure 2 shows the developed Simulink model for the study. Table 1 shows the range of parameter variations used in the study.

The Simulated Study

The study attempts to understand the robustness of the solar PV microgrid to sudden input changes in solar radiation levels. Such a situation can arise due to a passing cloud, wherein the uneven translucence of the cloud can result in changing levels of solar radiation incident on the solar PV panels. As a working base, the change in irradiation levels at 1 second interval for a simulation of 6 seconds has been considered. A step change has been considered for a given time interval of 1 second to create a situation that can result in a higher level of input variation gradient as compared to an incline change. Fig 3. shows the changes in radiation used for the simulation period. Figure 4 shows the voltage variations at the point of common coupling (PCC) of the solar PV section and the rest of the microgrid. A load of 40 kVA at the microgrid has been considered. The performance for the frequency transients in the simulation period was studied under three different power factors, namely, 0.8 p.f lagging, unity p.f and 0.8 p.f. leading.

RESULT OF THE STUDY

Case [A] Transient performance at 0.8 p.f lagging

Figure 5 shows the frequency transients for the simulation period when the load power factor is 0.8 lagging.

Case [B] Transient performance at 1.0 unity p.f

Figure 7 shows the frequency transients for the simulation period when the load power factor is 1.0 (unity)

Case [C] Transient performance at 0.8 p.f leading

Figure 9 shows the frequency transients for the simulation period when the load power factor is 0.8 leading.

DISCUSSION OF THE RESULTS

In all the three cases with different power factors magnitudes and nature, it has been seen from the simulations shown in figures 5-10, that the peak overshoots rise to a maximum of 50.225 Hz and can fall to a minimum of 49.93 Hz. Corresponding changes in the $\Delta f/\Delta R$ magnitudes vary from +0.5 to -0.5. This will probably imply that the frequency transients in the solar PV microgrid at modest power rating of 40 kVA will not be severe at power factors ranging from 0.8 lagging, unity or 0.8 leading. The study may be extended to solar PV microgrids of higher ratings (beyond 100-200 kVA) to examine if similar trends exist.

CONCLUSION

The paper attempts a study on the frequency transients observed for three different power factors, namely, 0.8 lagging, unity and 0.8 leading for a 40 kVA microgrid load fed from a solar PV systems with MPPT and inverter.

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The study indicates that the transients obtained for solar radiation changes from 1000 W/m² to 400 W/m² in step falls of 200 W/m² are within a maximum over shoot of 50.225 Hz, while the maximum undershoot is 49.93 Hz. The gradient of the frequency change to the radiation level changes also indicates a variation of ± 0.5 Hz/ W/m². It is felt that in the light of these simulated results, the severity of frequency transients in the system under step changes of solar radiation is marginal. However, it would be worthwhile to extend the study for higher rates solar PV systems to see if similar trends are obtainable.

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Table 1 shows the range of parameter variations used in the study.

Solar Irradiance Level (w/m²)	PF	Phase Voltage Varaition Range	Connected Load at Microgrid
400	0.8 lag	90-250 V	40 kW
600	1		
800	0.8		
1000	-		
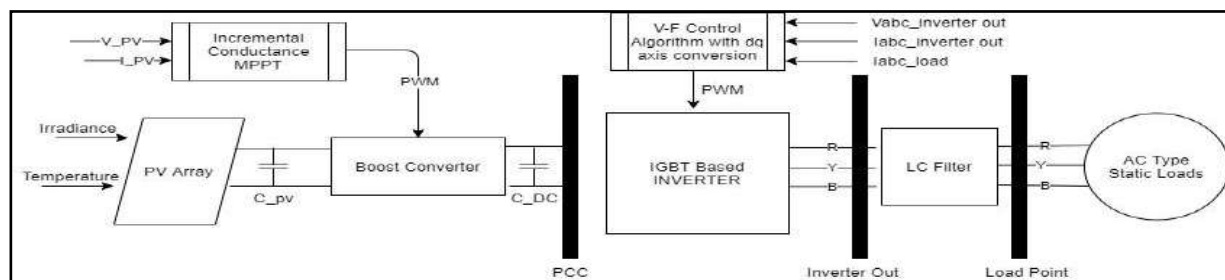
Fig. 1. Basic PV Microgrid System

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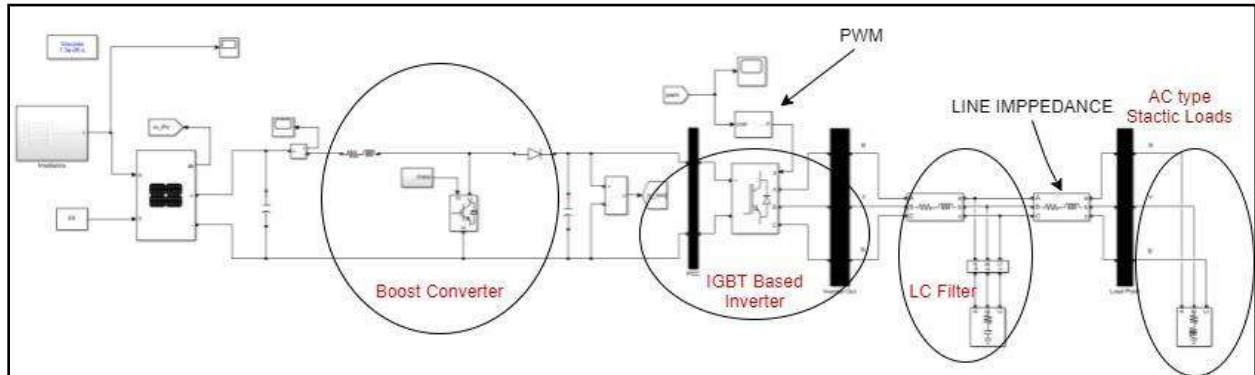
Fig. 2. Simulink model for the PV Microgrid System
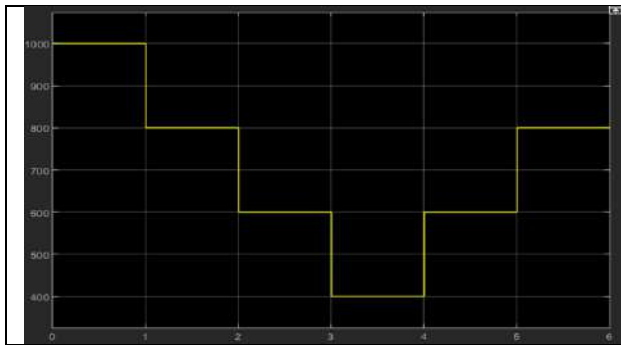
Fig. 3. Changes in radiation used for the simulation period	Fig. 4. PCC Voltage variation with irradiance change
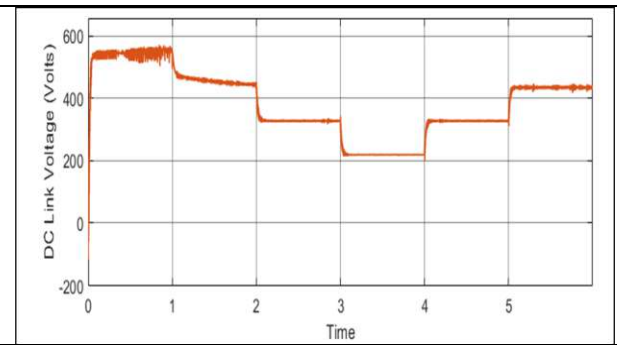
Fig 5: Frequency transients at 0.8 lagging power factor
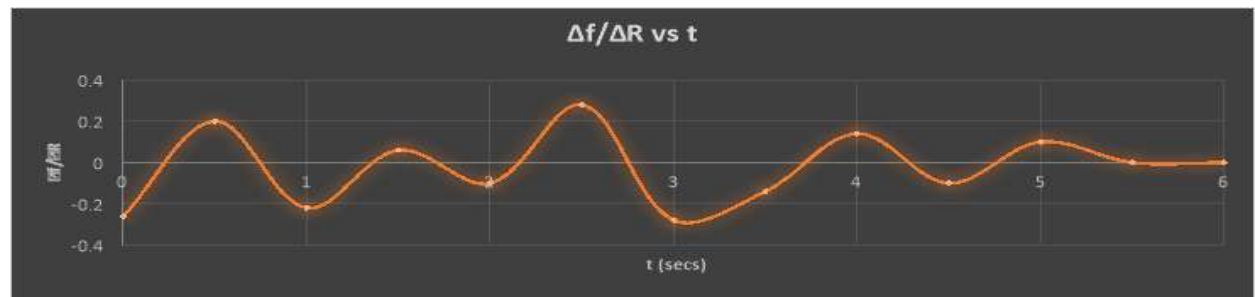
Fig 6: Frequency – radiation gradient (Δf/ΔR) as a function of the time period (t) for 0.8 lagging p.f.
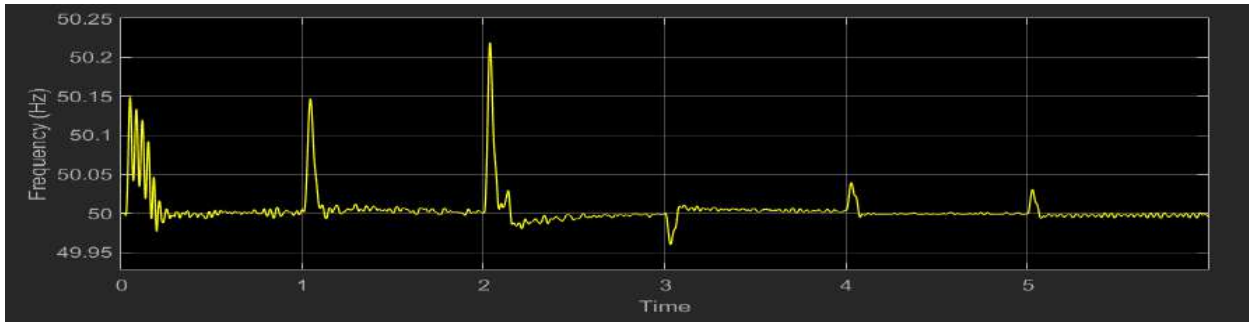
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Figure 7 shows the frequency transients for the simulation period when the load power factor is 1.0 (unity)
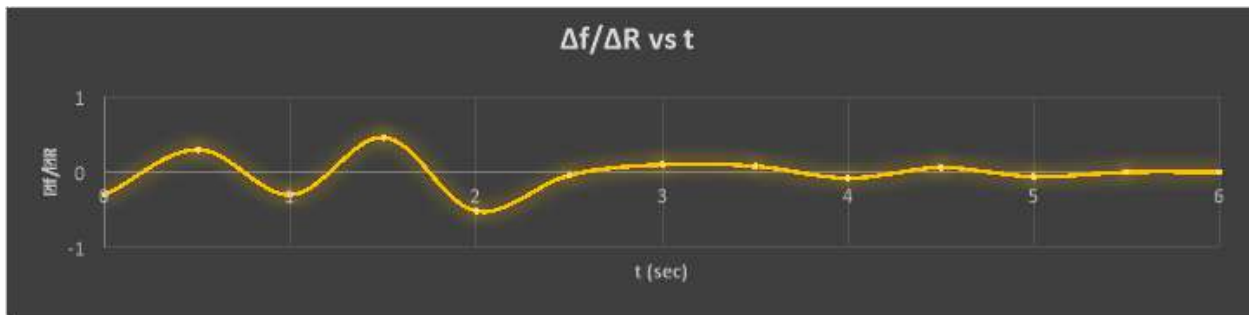
Fig 8 : Frequency –radiation gradient (Δf/ΔR) as a function of the time period (t) for unity p.f.
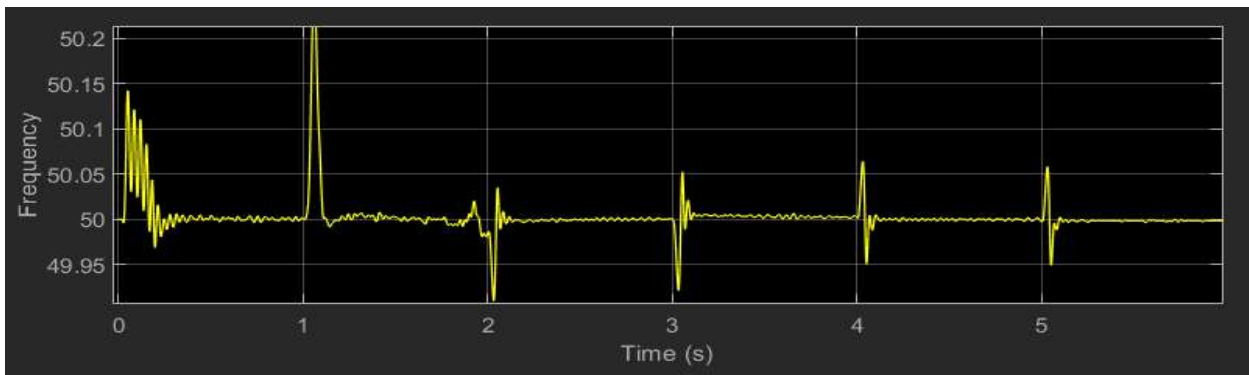
Fig. 9. Frequency transients at 0.8 leading power factor
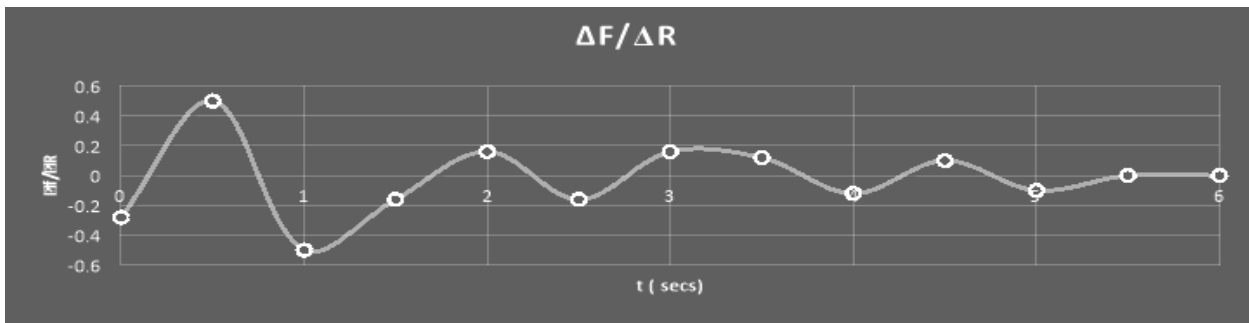
Fig 10: Frequency-radiation gradient (Δf/ΔR) as a function of the time period(t)for 0.8 leading p.f.

Trust Built BTG Access Controller Model in Wireless Sensor Network to Ensure Secured Data Transmission

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ABSTRACT

E-health monitoring system is increased in its popularity by providing flexible environment for the hospitals for storing their patient's health status information without keeping individual servers on hospitals. Information stored in the servers is sensitive information of patients which cannot be provided with public access. Usually the doctors are only allowed with access permission to operate the information whenever it is required. Thus it is vital important to doctor's presence to have approach to the patient's health information. However in emergency situations, in case of absence of doctors it is more difficult to retrieve the patient's information which would affect the treatment provided to the patients. And also in the previous work, BTG policy rules are defined already, in which emergency acts cannot be taken if none of them defined in the rules are not present. These problematic remains focused in our previous investigation method by leading technique called Resource Constrained Secured Access Control Module (RCSACM). In this work, the hackers might attempt to steal the data by acting as genuine users in the emergency act by performing identity foraging attack. And also transferring entire data through multiple data path links would cause increased delay and more bandwidth consumption. These problems are fixed in the proposed investigation method by familiarizing the new method namely Trust built BTG Access Controller Model (TBTGACM).

Keywords: Wireless sensor network, BTG policy, RCSACM, TBTGACM

INTRODUCTION

The foremost role for assigning a wireless - sensor networks: (WSNs) remains to screen the somatic world and give perceptions to different applications. As WSNs are typically conveyed in a situation that is helpless against numerous security assaults, it is spirited to resistors the entry to the wireless sensor formative nodes (Eg, perusing sensors information), particularly when around remain numerous clients in it. Also, unique clients may have distinctive access benefits. For instance, on account of WSN sent in a combat zone, fighter just needs to occured the information identified with his assignment, however a higher rank officer frequently requires data gathering for a general move and subsequently ought to have more data get to benefits than a warrior. The application will be endangered if get to control isn't appropriately authorized.

Access control can be executed by two methodologies, in particular, centralized and distributed [6]. A centralized access control approach requires a base station to be included at whatever point a client solicitations to get confirmed and get-to the data put away in sensors nodes. Outrageously, it is wasteful, not adaptable, and vulnerable against numerous potential attacks towards the extended communication path. For instance, for sensor systems conveyed in outrageous and unsafe situations, for example, seas and creature living spaces, it might be outlandish or restrictive to keep up a steady correspondence association between a network base station and the outside networks. In this mode, a centralized approach makes sense well just for little, trial networks, yet not for substantial scale sensor systems. Then again, in distributed access control, the authorized clients can enter the sensor-field to straightforwardly get to information on sensor hubs without including a base station. This approach can keep away from shortcomings, for example, single point of failure, execution bottleneck, which are unavoidable in the centralized case. These favorable circumstances together have prompted late expanding popularity of distributed information access control.

Trust Built Access Control Limitation Method

The primary goal of the research method is to ensure the successful data access control limitation thus the emergency cases can be treated immediately. This is achieved in this research method by introducing the Trust built BTG Access Controller Model (TBTGACM). In the planned work, B-T-G access permission is taken dynamically based on their trust value which is calculated using the previous transaction. In order to eliminate the more bandwidth consumption, data aggregation statics is performed instead of sending all sensor data in-separate route. This key objective of the proposed research technique is that to provide guaranteed access control to the manipulators in case of emergency act with the concern of trust and security issues.

System Architect

The traditional access controller display Resource BAC has pulled in the considerable measure of enthusiasm for some regions. The principle stuffs of this technique are the compulsion of pre-defined parts to the clients. Clients with the various parts might have diverse benefits, which could give system a fine grained control. Be that as May, it was unsatisfactory for the dispersed framework particularly WSN's. In proposed work, a Trust built BTG Access Controller Model is represented in interpretations based of the Resource BAC. The essential system of this ideal model is exhibited in the Figure 1. Primary thought about the presentation related to the trusts and the significance degree characteristics in foremost to the TBTGACM technique to create it handy for the WSN's.

Some real-fragments of access control system are depicted as takes after:

Administrator(A) - Substances that incorporate requirements to alter arrangement of authorizations, reliance levels, confidence strategies and client task.

Permissions (PE) - A portrayal of approved collaborations that decide if admittance possibly allowed. The significances of consents are nourished returned towards administrator, empowering dynamic change of limitations system.

Security Policy (SP) - An arrangement rules that used to confine the safekeeping chance. It's a piece of consents contributions.

Trust Level (TL) - The level esteems that quantity the dependability related to node. The trust is additionally a bit of contribution to the consents. It is related with particular parts.

Privilege (PR) - The privileges affirmed in system, which was identified with the client's part.

Risk Functions (RFs) - A capacity assesses hazard factor of another landing client.

Trust Evaluations (TE) - The Equable procedure which assesses the client's trust.

Trust Evaluation

Trust assessment related towards the greater portion of previous-effort for the greatest part centers is important dispute of dependence administration pattern. To appraise node's confidence, we intended to proposal, a confidence strategy. The dependence is calculated out of Direct trust and Indirect trust. In previous likewise remain partitioned into 2 units: most recent perception information part and the trust history information part that will stores trust record between the sensor-node in the predetermined organization, whereas the last remains gotten since suggestions of different nodes. Trust assessment processes can be spoken to as takes after.

$$T(i1,j1)_l = \alpha1 \times DT(i1,j1)_l + \beta l \times \frac{\Sigma_{(k1 \in N_j, k1 \neq i)} IT(k1,j1)_l}{|N_j| - 1} \qquad (1)$$

with $\alpha1 + \beta1 = 1$, $\alpha1 > 0$, $\beta1 > 0$. As in Eq.(1), T (i1, j1) denotes the dependence estimation of the device j1 for the node i1. Vertex i1 quantifies the confidence amid the nodes j1 in opinion of together the Directed trust- DT (i1, j1), and Indirected trust - IT (k1, j1). The amount l1 speaks to succession number of most recent assessment record. To the opinion of the Indirected trust, Nj is the customary of gathering all the nearby vertex of node j1. The resolution is to preferred the Indirected trust esteem gave nearby vertices are the furthermost cruel conduct could remain recognized by neighbor nodes.

Amount $\alpha1$, $\beta1$ were been measured components to be interconnected with the employment of the access controller policy. Parameters $\alpha1 > \beta1$ which illustrate that nodes in system is further persuaded about the judgment. Though locale $\alpha1 < \beta1$ shows that wants be allude to the recommendation by other further than themselves in the trust assessment. The expectation esteem is limited as $0 \leq Trust \leq 1$ where a advanced esteem demonstrates the higher trust worthiness. The valuation of the Directed Trust formula is calculated by:

$$DT(i1,j1)_l = \gamma * DT(i1,j1)_{l-1} + E(i1,j1)_l , \gamma1 \geq 0 \qquad (2)$$

Where, DT (i1, j1)$^{l-1}$ denotes to immediate trust esteem in the clarification of earlier recital of the vertex. The E (i1, j1)^l speaks to present deportment of the arrangement. The measured variables $\gamma1$ is to the exponent falloff stint to the factor, which measured using:

$$\gamma1 = e^{-\varrho*(t_c - t_{c-1})} ; \ t_c > t_{c-1} \geq 0, \varrho \geq 0 \qquad (3)$$

Where, t_c remains to the present time and t_{c-1} denotes the time duration, when the previous association happened. As mentioned foremost by the Eq. (3), trust esteem diminishes with elapse by of the given time. The load elements ought to-rely upon the unique situation. At the fact when $\gamma1 \ll 1$, it implies that significances for ongoing associations is significantly most critical relates that of more established one. The capacity E (i1, j1)$_l$is illustrate by:

$$E(i1,j1)_l = \begin{cases} P(a), & \text{for } (0 \leq p(a) < 1) \\ N(a), & \text{for}(-1 \leq n(a) < 0) \end{cases} \qquad (4)$$

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Here, P (∗), N (∗) speak-to the confident and unconfident evaluation for the node activity a separately. That 2 parameters should to take after the basic practice that a decent notoriety is more hard to select up than an awful one. At last, the Indirect Trust assessment procedure is figured using:

$$\sum_{k \in N_j, k \neq i} IT(k1, j1)_l = \sum_{k \in N_j, k \neq i} Dt \times Dt(k1, j1)_l \qquad (5)$$

As given, the indirected trust evaluation of vertex j1 for node i1 is processed allied-to the recommendation from neighbor of vertex j1. Regarding this technique, consumption of Trust based sequence to estimates the Indirect Trust of nodes.

Algorithm: To Compute- Node Trust.
Beginning state: Nodes needs-to link with added nodes in scheme.
Input: Nodes id, Security parameter, Reliability measure, Mobility Technique relates the nodes.
Output: Trust's esteem computation, correspondence.

Start:
Starting trust node measurement:
T1 initial= (S+U)/(T1 i+S) or Pr1;
If (T1 initial is adequate for the communications)
Permit correspondence with nodes.
else measure the dependence security display for nodes.
T1s =A+ E + R;
If (T's is correspondence satisfactory)
At that argument permit correspondence with nodes.
else measure Trust an incentive of versatility show for node.
If noe is fixed
In this point expects the trust estimation regards nodes in versatility demonstrate is 0.
else measure the Trust worth towards- the nodes was versatility.
T1m = Me + Em;
If (T1m is correspondence satisfactory)
It permits the correspondence with the nodes.
else find Trust on behalf of the nodes-in Reliability Model.
T1r =D + Ed;
If (T1r correspondence sufficient)
It will permit correspondence;
Else compute the general node Trust.
Overall Trust = T1 initial + Ts + Tm + Tr;
If the Overall Trust is fulfill for correspondence
They will permit the correspondence nodes.
else
deny the correspondence with nodes.
end
Notations:
T1 initial= Trust Initial value scheming.
S = Effective interaction.
U = Failed interaction.
Ti =Total number of Interactions
Pr = Peer node recommendations.
Ts = Security model - Trust metric of its nodes.

A = Admission control.
E = Packets Encryption.
R = Routing of the secured packets.
Tm = Trust Evaluation belongsto node in flexibility model
ME = Estimation of the mobility.,
Em = Energy Consumptions during the node mobility model.
Tr = Trust values of nodesin-consistency models.
D = Node Data's fusion.
Ed = Data fusion Energy.

Trust Based Access Control Limitation

The Trust, exposure factors are urgent parameter to characterize when node was adequate. Higher trust esteem some nodes that makes simpler when that was joining the system. Essentially, the vertex had a lesser chance, which will in all probability be conceded admission privileges. In these model, remote nodes isn't allowed to by-enter the system except if both two properties fulfill the prerequisites. Not every individual of node in system have benefit for enable the remote nodes to join in predefined formation. Contingent upon unique situation, that benefit is established by the head. Furthermore, TBTGACM display an adaptable access controller models. It isn't plan intended at the sensor related networks deprived of central CA for approval, yet additionally a discretionary strategy for the unique that en-counts the entire validation framework. On the off coincidental that another entry sensors nodes have the key join, that will acheive a high trusted degree promptly. Procedure for other landing nodes to join the system functions as takes after:

Step 1: The recently arrive node named N sends the demand to terminus nodes. About this ideal, the admission demand for is the four - ary tuples, more-over it is meant as $U_r = <U_{id}, R, T, Key_{join}>$, wherever u_{id} represent the basis nodes IDs, R is part initialize the nodes solicitations toward actuate, and t is the time-stamp. Besides, demand nodes may incorporate the key node join in the occasion that intakes one.

Step 2: At the point when the endpoint node gets the requests it can checks whether it takes the factual to permit the innovative node to intersection the system. On the off chance that takes, it resolve figure of immediate confidence estimation of the fresh hub and it directs the trust demand to the nearby neighbor of the novel nodes aimed at acquiring the suggestions. The nearby neighbors list that be refreshed thru the current neighbor disclosure processes, for example- the communications of the periodical hi packet.

Step 3: Vertices gets confidence demand to identify, whether-node is asked for objects. In the occurrence that, they will send trust answer. Else, the nodes spirit basically stays quiet.

Step 4: For the wake of acquiring the suggestions, the destination vertex utilizes the resident trust based assessment procedure to figure out trusted estimation linked to fresh debut nodes-through the Equation (1), which was stated before. What's more, the novel vertex that associated to the key -join related to consider managing an extraordinary confidence levels, when that nodes have no previous records in system. Notwithstanding, the key-join should be void, when node picks up a bad reputation in the pre-existing system. Related this will keep traded off nodes from propelling an inside assault. In point, when the new vertex enters the trusted assessment processes, hazard appraisal assumes control to contemplate the danger of the regulator.

Step 5: Endpoint nodes forced, to choose-related about to offer consent near the new coming node N. The destinations node might be ruined; it is perilous, when the choice to provide or not to consent to another landing node to bond the arrangement is completed by just one single node. By the overhead procedure, the fresh arrival nodes would approach system and to acquire comparing benefits while it gets in excess of 2 certificates as of various destination nodes.

EXPERIMENTAL APPROACH RESULTS

In contemporary implemented segment, the execution estimation of whole research effort stays completed in the NS2-simulation atmosphere alongside execution measurements for combining both the pre-standing and future frameworks. The current effort will give low concert; however, the performance routine is boosted-in the projected framework by bringing in different fresh calculations. Examination assessment is done among the current and projected frameworks to be specific BTG-RBAC, BTG-AC, IBTG-ACMS (Previous work), RCSACM (past work) and Trust aware Break the Glass Access Control Model (TBTGACM). The setting esteems introduced aimed atnetwork-configurations amid the examination that are specified in the Table I. That quality could be shifted & efficient aimed at various presentations. In current instance of exploration, the estimations of period interims in Table-I that specify related to lessen the experiments duration for the reflection. Security Level: The Confidence parameter of this frameworks in such-way of complete the information communication is detailed by the lessened data packets loss / corruption frequency. The imitation measures esteems got sophisticated security level metric is appeared-in the accompanying table 2.

From figure 2, demonstrate that the projected-technique prompts give the better outcome to the extent that enhanced security level. The proposed practices has expanded security execution than the current exploration strategies by guaranteeing the secured access control instrument which confines the malevolent clients from getting to patient's medicinal services data. From this investigation it demonstrated that the anticipated technique TBTGACM has expanded security level where it was 25% higher than RCSACM, 73% higher than IBTG-ACMS, 112% higher than BTG-AC and 197% higher than BTG-RBAC   Energy consumption: Energy utilization is considered, as the measurements of energy devoured by the whole system for finishing information transmission effectively. The simulation measure esteems acquired for the energy c utilization metric is appeared in the accompanying table 3.
Figure:3 represents towards the appraisal that offered BTG-RBAC, BTG-AC, IBTG-ACMS, RCSACM and TBTGACM approaches using the foremost method figure for energy-consumption. The development variableranges from 20 to 60 (s). By this investigation we can be demonstrated that projected technique TBTGACM has lesser energy utilization where it was 32% lesser than RCSACM, 43% lesser than IBTG-ACMS, 60% lesser than BTG-AC and 65% lesser than BTG-RBAC.

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Table I: Parameter metric for the Experiment

Parameters	Values	Units	Descriptions
Nu	50	Sensor Nodes	Number of implemented Nodes
Cl	4	Cluster	Total quantity of cluster formed
$T_{re-cluster}$	40000	Mille second	Sample time to re-cluster
T_{sample}	600	Mille second	Mockup Time for Sensing
T_{cycle}	6000	Mille second	data transmission time interval
$T_{Data-Rx}$	400	Mille second	Cluster Head receiving data time
$T_{Data-agg}$	60	Mille second	CH data aggregation time
$T_{Radioon-CH}$	700	Mille second	Determined duration time to keep radio-on for transmission
$T_{Radioon-CM}$	200	Mille second	Determined duration to keep radio on for transmission
$\Delta-V_{th}$	200	Mega volt	Voltage-threshold for the departed node

Table. 2. Security level comparison values

Simulation time	Methods				
	BTG-RBAC	BTG-AC	IBTG-ACMS	RCSACM	TBTGACM
5	0.07	0.15	0.23	0.35	0.41
10	0.14	0.3	0.46	0.69	0.85
15	0.2	0.4	0.49	0.715	0.92
20	0.28	0.48	0.52	0.755	1
25	0.4	0.56	0.65	0.86	1.1
30	0.5	0.63	0.77	0.97	1.2
35	0.57	0.69	0.81	1.09	1.35
40	0.64	0.73	0.87	1.2	1.49
AVERAGE	0.35	0.49	0.6	0.83	1.04

Table. 3: Energy-consumption comparison values

Simulation time	Methods				
	BTG-RBAC	BTG-AC	IBTG-ACMS	RCSACM	TBTGACM
10	0.53	0.47	0.19	0.12	0.07
20	0.89	0.8	0.62	0.48	0.32
30	1.11	1.01	0.78	0.695	0.43
40	1.36	1.17	0.82	0.7	0.52
AVERAGE	0.97	0.86	0.6	0.5	0.34

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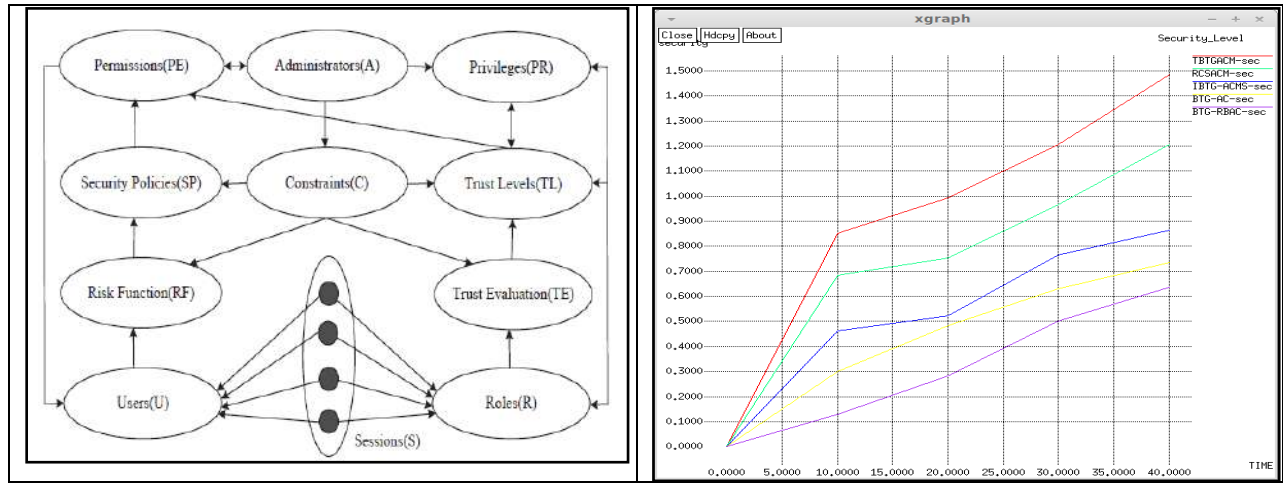
Figure 1. Basic Architect of TBTGACM model
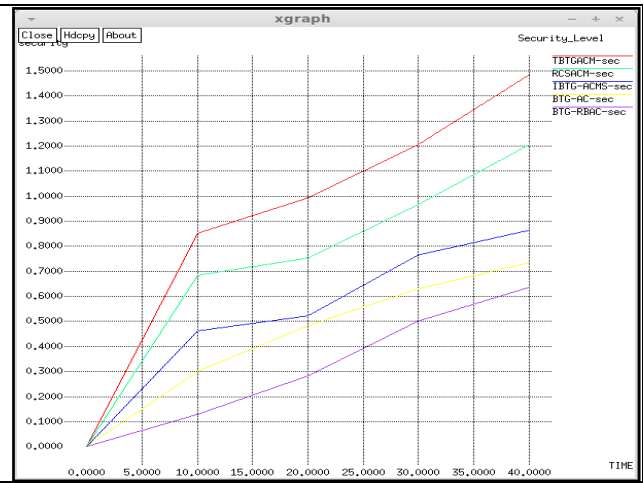
Figure 2. Security level ratio
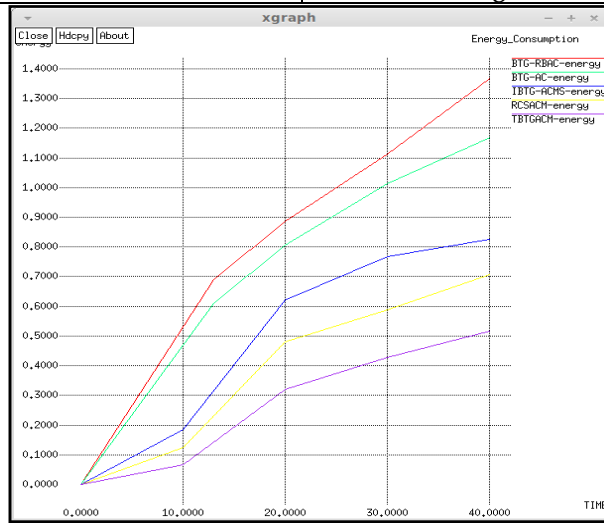
Figure 3: Energy-Consumption ratio

RESEARCH ARTICLE

An Improved Recommendation System Based on Hybrid Data Mining Technique

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ABSTRACT

the recommendation system is an essential application in supporting to understand the patterns according to the behavior. This technique can also be helpful for finding a solution for complex problems where similar solutions are used in various problematic situations. In this context, this paper is concentrated on the study of recommendation systems. Thus first an in depth analysis is conducted on existing methods and tools. In addition to a review of recently contributed approaches is reported in this paper. Further, a data mining model based on concluded techniques and algorithms is reported here. That technique usages a matrix of M attributes and N solutions as a dataset. Next, the FCM (fuzzy c means) clustering and Hidden Markov Model (HMM) is implemented for predicting the most suitable solution for the currently given problem. The experiments are conducted with a different set of data instances and the model's performance matrix is provided. the obtained experimental study demonstrates the proposed hybrid recommendation model is superior then the classical content-based recommendation model and Apriori algorithm based market basket analysis system. Finally, the conclusion and the future research direction are reported in this paper.

Keywords: recommendation system, data mining, prediction, clustering, data analysis, pattern recovery;

INTRODUCTION

The recommendation system is a method for finding the appropriate services and products among similar available. The recommendation systems are initially developed to support e-commerce platforms, which contains a significant amount of products as well as users [1]. The recommendation systems help to locate the fit products according to the habits, behavior, need and/or other factors. This technique is further extended to making it acceptable for other application areas such as caching, video recommendation, health industry, tour and travels industry, education and many more [2]. Based on the application areas the design and implementation may differ from each other. The recommendation systems are designed using data mining techniques. Those techniques are helping to explore and analyze the historical data and obtain valuable patterns for future use or recommendation. According to the study, this technique not only focused on some specific kinds of data mining algorithms [3]. These techniques are usages the techniques of association rule mining, clustering, optimization, classification and other kinds of specifically designed algorithms [4]. In this work, we are focused on designing and developing a new kind of recommendation system which accepts the N number of different generalized problem attributes and produces the M number of suitable solutions according to their fitness.

In this context, the proposed work first includes the investigation about the different concepts and methods which are used for designing the recommendation system. in the next step, we are exploring different techniques contributed by different researchers for improving the recommendation system for different areas of application. Further, a recommendation model is proposed for design and development using the data mining technique and their performance analysis is reported with respect to frequently used recommendation models. Finally, the conclusion is provided and the future road map on research work is also reported.

BACKGROUND
This section offers the basic concepts of recommendation system design and used algorithms for designing the proposed system.

Recommendation System
The recommendation systems are a kind of system, which can recommend products, items or services and most likely according to the specific user interest. Here we are giving an explanation of the recommendation system and significance. Recommender systems have become an important area recently. The significant amount of content on the web and other databases makes the recommender system an essential part of the digital world. It tries to direct users to where they would like to go without losing specific patterns in the huge amounts of information. That is currently used for recommending books, CDs and other products. Amazon.com [5] is an example of such a system. Here web-page has proved a valuable means of Web users by providing useful and effective suggestions. The core techniques in the recommendation are the learning and prediction models that learn on users' behavior and evaluate what users would like to view. It can suggest interesting items from a large set of items based on the knowledge gained. A Good recommendation can improve user satisfaction [6].

Through the continuous development of internet data, it has become a huge repository of information and keeps growing exponentially. However, the human capacity of reading, accessing and understanding is limited. Therefore, it is complicated for Website administrators to supply the contents according to the individual user's need. Therefore the behavior analysis is useful to understand the requirements of users. In this context the personalization of online services are required [7]. The recommendation is a popular technique and demonstrates the connections among stories, products, or services for a website. That is a type of data processing model which tries to predict the user's next activity then recommends items which are most suitable according to user. It is a web mining based solution used with e-commerce. According to the consumer's expectation RS are providing ease in discovering products likely to purchase. Thus it is useful in eCommerce sites that offer millions of products [8].

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Categories of Recommendation System

There are a number of techniques for Recommender System design, and each of them have it's own potentials and limitations, which make them suitable for applications. Figure 1 provide the relationship between different techniques. A RS can be implemented using number manners. According to their implementation aspects we can categorize them in the following manner.

- **Content-based:** Similar items to the specific user's preference in historical record and used as recommendation
- **Collaborative:** Items are suitable for the users who have a similar behavior of users are generated as a recommendation.
- **Hybrid Approach:** This approach is used for combining the above given methods into a common platform.

Content-Based Methods: In content-based recommendation method, the rating of an item R for user C is estimated based on the user rating compared to the other items that are similar to S. For an instance in a movie recommendation, a user is recommended a movie according to the movies that she/he rated highly in the past. The movie that has the highest similarity to the preferred movies of the user in the past would be recommended. There are several methods of finding the similarity of an item to a set of other items. The keyword analysis techniques can be applied to find recommendations. The algorithm analyzes the content of the previously preferred and rated items for each word in the item and measures the closeness that is not seen by the user. One of the best-known techniques is the term frequency/inverse document frequency measure [9].

Collaborative Methods: these techniques are working with the user interest and product ratings not only for specific user's preference and/or rating. An appropriate example of this technique is the book and CD RS [10]. Collaborative algorithms can be two types: model-based or memory-based. This approach also considers the behavior of similar users. Therefore the similar users have found to produce recommendations. This task can be problematic for new users since they do not produce ratings to enable the finding of similar users. New items are not easily recommended to the users in collaborative filtering since the item is not rated by a substantial number of users, the new item will not be recommended to anyone. Another limitation of the collaborative filter is the number of rated items. Generally, the number of rated items is relatively low when compared to the number of items to be rated. Therefore an effective system is needed, which gives accurate ratings based on little underlying data [11].

Hybrid Method: Such a recommendation aims to avoid certain limitations of filtering methods by combining two or more methods. The hybrid approach combines the contentbased and collaborative algorithms to make it [12]. Different methods are used to combine both the algorithms:
- Developing the content-based and collaborative technique separately, and combine the recommendation of both the techniques.
- Implementing the features of collaborative filter into content-based technique.
- Or applying the content based technique over the collaborative algorithm.
- Or combining the qualities of both the techniques into a common framework.

Applications of Recommendations

There is various application of recommendation systems, several important applications of this systems are as [13].
1. **Product Recommendations:** On of the most essential use of RS is e-commerce websites. The ecommerce platforms are providing recommendations to most of the returning user, which is likely to buy. This recommendation is done by the previous behavior of user and the similar user's purchasing decisions.
2. **Movie Recommendations:**Similarly Netflix is recommending the movies to their customers, which is might like by the user. That is performed based on user ratings. In this application accurate rating prediction is required, in this context; Netflix has offered a prize of one million dollars. The prize was won in 2009, by a team "Bellkor's Pragmatic Chaos".
3. **News Articles:** News services are trying to find user interest relevant articles, using the previous readings. The similar articles are obtained by using words in the previous readings.

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Frequent Pattern Mining

Frequent pattern mining has been an engaging subject in data mining research. Frequent patterns will be patterns that show up frequently in the data index. Finding frequent patterns assumes a job in mining associations, connections, and other fascinating connections. For instance, a lot of things as, glue and brush, which shows up frequently together in an exchange data index, are frequent itemset. Think about the situation, for example, purchasing a PC and afterward a pen drive, and on the off chance that this pattern happens frequently in shopping, that pattern is a frequent pattern. A substructure can represent various structures, for example, sub-graphs, sub-trees, or sub-lattices, which might be joined with item-sets. Frequent pattern-mining scans for repeating connections in a given information collection. Market basket investigation is the soonest type of frequent pattern mining. Frequent pattern mining was proposed in [14] for market basket examination. It examines purchasing propensities by discovering associations between the diverse items that clients put in their "shopping carts". For example, if clients are purchasing milk, how likely would they say they are going to purchase grain on similar outings. Such data can prompt expanded deals by helping retailers do specific advertising their products. Since the primary proposition of this new data mining task and its related mining calculations, there have been many follow-up research distributions, on different sorts of applications, going from versatile data mining strategies to dealing with a wide variety of data, and new applications. Over a time of productive research, the time has come to play out a review of this field and analyze what more to be done so this innovation a foundation of approach in data mining applications [15]. That is a wide zone of research, and it identifies a wide subject from an application viewpoint. The examination in the zone falls into four classes:

- **Technique-Centered:** This identifies with the assurance of progressively proficient calculations for frequent pattern mining. A number of techniques are available which are utilizing different type of tree based techniques, and distinctive data strategies notwithstanding that, various varieties, for example, the assurance of compacted patterns of incredible enthusiasm to specialists in data mining.
- **Scalability Issues:** The frequent pattern mining techniques are suffering from critical scalability issues. At the point when the data touches base as a stream, multi-pass strategies can never be utilized. When the data is vast, at that point parallel information structures must be utilized.
- **Advanced Data Types:** Various varieties of frequent pattern mining have been proposed for cutting edge data types. These varieties have been used in a wide assortment of assignments. The unique information spaces, for example, image data, tree-based data, and gushing information regularly require uncommon calculations for frequent pattern mining. Issues of intriguing quality of the patterns are very important in this setting [16].
- **Applications:** Frequent pattern mining has various applications to other information mining issues, Web applications, programming bug analysis, and synthetic and natural applications. A lot of research has been dedicated to applications on the grounds that these are especially essential with regards to frequent pattern mining.
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FCM (fuzzy c means)

The aim of the FCM (Fuzzy C-Means) clustering is to split the data into N number of groups. It is an unsupervised learning technique for categorizing the data. In this context there is a vector which contains data instances $V = \{x_1, x_2, x_3, \ldots, x_n\}$ [17]. Where x_i is the individual instance of data. Now if we need to create k number of data segments then we need a distance or similarity function to make decisions of clusters. In FCM algorithm a membership function is used. Thus the assigned categories are $C = c_1, c_2, \ldots c_k$. Here the membership function is defined as:

$$M_{i,j} = \frac{1}{\sum_{k=1}^{C} \left(\frac{\|x_i - c_i\|}{\|x_i - c_k\|} \right)^{\frac{2}{n-1}}} \ldots \ldots \ldots (1)$$

The membership function provides the bounding among data and the centroid. Using this function the initial decision is made to assign a label to the data. But for deciding the stopping criteria an objective function is required.

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When the improvement in objective function is tends to zero then the algorithm stop working. Therefore the following equation is used as objective function.

$$O_n = \sum_{i=1}^{N} \sum_{j=1}^{k} M_{i,j}^{j} \left\| x_i - c_j \right\|^2 \dots\dots\dots\dots (2)$$

Where, O_n is the objective function, $M_{i,j}^{j}$ is membership function used to establish the relationship, finally $\left\| x_i - c_j \right\|^2$ is a distance function namely Euclidean distance. However the FCM (fuzzy c-means) algorithm is suitable for segmentation and cluster analysis.

Hidden Markov Model

An HMM is a two hierarchy level process, which is used to model complex processes. In this process an observation is prepared based on related probability distribution. The HMM is defined in the following manner [18]:

Let there are N number of states, which are denoted using $S = \{S_1, S_2, \ldots, S_N\}$. A state at time t is represented as q_t. Similarly, let there are M number of observations per state and can be denote as $V = \{V_1, V_2, \ldots, V_M\}$. The state transition probability matrix A can be defined as:

$$A = [a_{ij}]$$

$$a_{ij} = P(q_{t+1} = S_j | q_t = S_i), 1 \le i, j \le N; t = 1,2 \dots$$

Here $a_{ij} > 0$ for all i, j.

$$\sum_{j=1}^{N} a_{ij} = 1, 1 \le i \le N$$

The probability matrix $B = \{b_j(k)\}$, where

$$b_j(k) = P(V_k | S_j), 1 \le j \le N, 1 \le k \le M \text{ and}$$

$$\sum_{k=1}^{M} b_j(k) = 1, 1 \le j \le N$$

The state probability vector r= πi, where

$$\pi_i = P(q_1 = S_i), 1 \le i \le N$$

Such that

$$\sum_{i=1}^{N} \pi_i = 1$$

The observations $O = O_1; O_2; O_3; \dots O_R$, where O_t is one of the symbols from V, and R is the number of observations. A complete detail of HMM requires to measure two key parameters, N and M, and three probabilities A, B, and π. The notation $\wedge = (A; B; \pi)$ is used to define a set of parameters, where A, B contain N and M. An observation series O, can be generated by possible states. For a sequence $Q = q_1, q_2, \dots q_R$, where q_1 is the initial state. The probability of O is computed from states given as:

$$P(O|Q, \lambda) = \prod_{t=1}^{R} P(O_t | q_t, \lambda)$$

Where, independence of observations is expanded as:

$$P(O|Q,\lambda) = b_{q1}(O_1)b_{q2}(O_2)\ldots\ldots b_{qR}(O_R)$$

The probability of Q can be describe as,

$$P(Q|\lambda) = \pi_{q1}a_{q1q2}a_{q2q3}\ldots\ldots a_{qR-1qR}$$

The observation probability O can also write as:

$$P(O|\lambda) = \sum_{all\ Q} P(O|Q,\lambda)\,P(Q|\lambda)$$

The computation of $P(O|\lambda)$ using the procedure which is known as Forward-Backward technique.

LITERATURE SURVEY

This section includes different techniques recently presented to improve the performance of the existing recommendation engine. The recommender systems (RS) used to extract relevant information from the data. Considering the historical limitations, Context-Aware RS are deployed, which leverage contextual information. *K. Haruna et al* [19] present a review of a recent development for the context-aware RS. This contributes by taking an approach to the complete CARS process. First, the review is presented and classified literature, considering the applications, filters, extraction and evaluation approaches. Second, viewpoints are presented relating to merit and demerit. Finally, the challenges and opportunities for the future are highlighted. In recent years, deep learning has garnered interest in many research fields. The influence of deep learning is also pervasive, it demonstrating effectiveness when applied to information retrieval and RS. *S. Zhang et al* [20] aims to provide a review of deep learning-based RS. They provide taxonomy of deep learning-based RS, along with a summary of the state of the art. Finally, they expand current trends and new perspectives to this new and exciting development. RS is the Matrix Completion setting: partially known matrix of ratings given by users (rows) to items (columns). Few attempts were done to handle that objective with Neural Networks, an architecture based on Auto-encoders proved to be promising. *F. Strub et al* [21] enhanced the model by managing the missing values computed using loss function, and also involving additional attributes. The results show that the additional features slightly enhancing the error, but, it have significance on cold start problems. It has been important to consider the trade-off between the accuracy and time in RS. *M. Nilashi et al* [22] develops a hybrid recommendation method based on Collaborative Filtering (CF). Authors solve two drawbacks, sparsity and scalability, using dimensionality reduction and ontology. They use ontology and SVD is used for enhancing the prediction accuracy, to find the most similar items and users. They significantly improve scalability. The results showed that the method is effective in improving sparsity and scalability.

RS are filtering tools to predict the rating for users and items. This system provides will assist the users for finding the similar interests and users. *R. Katarya et al* [23] objective is to develop a recommendation system using clustering and prediction technique. In this context they use k-means and cuckoo search techniques. The performance of model has measured for obtaining better results. The results obtained show that may provide high performance regarding reliability, efficiency and accurate personalized movies. RS deal with large spaces of items and needs to serve billions of users. *M. Chen et al* [24] present a recipe for such a production top-K RS at YouTube, with the REINFORCE algorithm. The contributions are: (1) enhancing the REINFORCE as a production recommender; (2) applying the off-policy correction to data biases in learning from feedback collected; (3) a novel top-K off-policy correction to recommending multiple items; (4) showcasing. To help authors for deciding to submit their articles, *D. Wang et al* [25] present a Journals & Conferences RS. This recommends appropriate journals or conferences using a priority value which is obtained by abstract of article. In order to update training set and also learning model a web crawler is used. Authors propose a hybrid model, developed using regression and chi-square feature selection. The results show it can recommend most suitable conferences and journals. The sales conversion can be improved based on RS,

which learns user preferences based on previous buying pattern and business metrics. Y. Sun et al [26] propose to integrate dialog systems and recommender into a deep reinforcement learning to build a personalized recommendation agent. They represent a user discussion history as a user query. A belief tracker is used this generated query and updated by natural language. The author proposes a set of actions tailored to recommendation agents and trains a deep network to decide which action should take. Authors train a model that uses both past ratings and query in the current session when making predictions. The system tries to combine user priorities by using question answering. When sufficient preference is obtained then the recommendation are generated. T. Schnabel et al [27] study interface component for RS, the aim is to support the user's decision, and feedback to improve recommendation. A shortlist of candidates is created from a cognitive perspective. That serves as short-term memory. A ML technique which is adding items to the list prepared using feedback of product exploration and decision making. Short listing provides data for training without the increased load that requesting feedback. The author performs a study with a movie recommendation to compare interfaces with those that do not. Using this study author has found: (i) a shortlist is very useful in decision making process; (ii) an interface and appropriate short listing support is also attracting the users, and (iii) in order to improve the quality of recommendation we need to include a feedback technique and a shortlist. RS based on collaborative and content-based methods working on user-profiles, tags and previous user preferences. Such methods are useful for solving cold-start problem, privacy, and contexts aware techniques. T. Osadchiy et al [28] describe a model for collective recommendation based on user personal interests which is simple in execution. The performance is evaluated on a realworld food processing system.

PROPOSED MODEL

This section explains the proposed work functional aspects and the required algorithm for providing the recommendation. The section first explains the system overview and then the system architecture is discussed. Finally, the required algorithm of the recommendation system is provided.

System overview

The recommendation system is a technique that provides the suggestion for selection of the most suitable solution among all the available solutions. In order to perform this, we need some data mining algorithms which analyze the historical data attributes and then required a prediction algorithm that offers the solutions. In this presented work a generalized recommendation system is proposed for design and implementation which investigates the historical data records based on FCM (fuzzy c means) clustering algorithm. This algorithm is also used to find similar data instances using which the observation and transactional matrix is developed for the hidden Markov model (HMM). The hidden Markov model is one of the complex prediction algorithms which is used the concept of the probability distribution for computing the upcoming event. In this presented work the main aim is to explore the technique of recommendation system design and their core concept understanding. That technique is basically further used for designing a recommendation system for the plant disease recovery solution development. In this context first, an image classification technique is used to identify the crop name, disease, and the stage of the disease. After obtaining these aspects of crops the recommendation model is used for predicting the most suitable solution based on the available solutions and their rating. Therefore in this presented work, the hybrid recommendation system is proposed for designing and development. The proposed model is usages the FCM and HMM algorithm for designing the required recommendation system.This section offers a basic overview of the proposed system for designing the recommendation system and the next section the proposed methodology is described.

Proposed methodology

The proposed methodology is demonstrated using figure 2. In this diagram, the required components and their functional components are described.

Input dataset: the dataset is an essential element of data mining and machine learning models. In this work, we are preparing a data model that is used to predict the next possible solution for the given problem. Therefore there are two datasets are considered in the format of the M X N matrix. Here the M number of observations and N number of solutions are included to develop such kind of data models.

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Dataset preprocessing: data preprocessing technique is a method by which the data is quality is improved using the different methods. These methods can include the data normalization, transforms, reduction, mapping, and others. In addition to that the preprocessing techniques are also responsible for reducing noise on data. Therefore irrelevant data, characters, data instances, and attributes are reduced. In this work, the missing values and incomplete data instances are reduced as the process of data preprocessing.

FCM: FCM (fuzzy c means) clustering is an unsupervised learning technique for categorizing data. The advantage of utilizing unsupervised learning techniques is that: these algorithms are efficient as compared to other learning techniques additionally it not required additional learning feedback for improving their correctness. These algorithms are working on the basis of the optimization model which is continuously improving self by satisfying the objective function. In this work, the dataset is subdivided into 4 clusters according to the included solutions in the dataset class labels.

Similar patterns: the outcome of the FCM clustering algorithm produces four clusters with similar patterns. Therefore these clusters are used for preparing the required matrix for making predictions or recommendations. These two matrixes for prediction are:

●**Observation matrix:** an example of observation matrix is given in table 1. This matrix contains the observational states and the states to define the observation matrix. This matrix shows the states are varying with the different patterns.

●**Transition matrix:** The transition matrix demonstrates the correspondence between states to state. The example of transition matrix is given using table 2

HMM: the hidden Markov model computes the probability of states based on the complex computed two above given matrix as given in table 1 and 2. in this function three inputs are required observation matrix, transition matrix and the current state for which the next states need to predict. That function is defined using the following equation:
$$\wedge = (A; B; \pi)$$

Recommendation: that is the final outcome which is computed for finding the next state probabilities. The higher probability states are recommended as the final outcome of the system, based on which the performance of the model is computed.

Proposed algorithm
The proposed data model described in the previous section is summarized in this section using the algorithms as given in table 3.

RESULTS ANALYSIS

This section offers the analysis of the results and provides a comparative study of the similar recommendation models. First, we have measured accuracy, in order to measure the accuracy we are utilizing a sequence as input, and tried to

predict sequence's next value. Based on predicted and known value the accuracy is measured. That can be calculated using:

$$accuracy = \frac{total\ correctly\ predicted\ data}{total\ data\ to\ be\ predict} X100$$

The accuracy is compared with two similar recent algorithms. The comparative performance is given in table 4 and their graphical represented in figure 3 (A). Here, X-axis represents the dataset size and Y-axis shows the accuracy of

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the algorithm. The accuracy is measured in percentage (%). According to the performance the proposed algorithm provides higher accuracy as compared to other two algorithms.

The error rate of a machine learning technique shows the amount of incorrectly predicted data instances. Therefore that is measured using the following formula

$$error\ rate = 100 - accuracy$$
Or
$$Error\ rate = \frac{total\ incorrectly\ predicted\ data}{total\ data\ to\ be\ predict} X100$$

The error rate of the recommendation systems are described in figure 3 (B) and table 4. In this diagram, the X-axis shows the dataset size and Y-axis shows the error rate of the algorithms. The error rate is measured in percentage (%). According, to the performance, the entire algorithm shows reducing error rate but the proposed model demonstrate the minimum as compared to both other algorithms. The memory usages indicate the amount of main memory acquired by algorithm. The memory usages can be measured using the following formula:

$$memory\ usages = total\ memory - free\ memory$$

The memory usages of the recommendation systems are reported in table 4. and line graph is given in figure 3 (C). The X-axis contains dataset size and Y-axis shows memory usages in KB (kilobytes). According to the performance in terms of memory usages, it is increasing with the size of data to be processed. But the proposed algorithm shows low resource consumption as compared to the other two algorithms. The total time taken to complete the data processing using the target algorithm is known as time consumption. In this context, the time requirements are measured using the following function.

$$time\ consumption = end\ time - start\ time$$

The proposed recommendation system is compared with two recent algorithms and performance in terms of time consumption is recorded in table 4. The values are represented using a line graph as given in figure 3 (D) in terms of milliseconds. To represent the line graph X-axis shows the size of the dataset and the Y-axis shows the time consumed. According to the performance of all the algorithms are increasing with the amount of data. But the proposed algorithm shows efficient performance as compared to the other two algorithms.

CONCLUSION

The aim of the proposed work is to study and analyze the techniques and methods for developing an efficient and accurate recommendation system. In this context, the paper includes the study of concepts and algorithms which are used for designing the recommendation system. Further, the two algorithms are addressed which are used for implementing the proposed recommendation system. Additionally to understand the problem-solving techniques using recommendation systems the recently developed and implemented techniques reported. Finally, the proposed model is described. The proposed recommendation system first includes a dataset of M X N size. In this dataset M number of attributes and the N number of samples are included. Further, the FCM (fuzzy c means) clustering algorithm is implemented for identifying the patterns associated with the dataset. The obtained patterns are organized in two matrices namely transition matrix and observation matrix. Further, these two matrices and the current state are produced into the HMM (Hidden Markov Model) for predicting the next states based on calculated probability distributions. The implemented technique is further evaluated for finding the performance of the system. This model is compared to the two popular techniques. Based on the comparative performance study the summary of performance is prepared as demonstrated in table 5. Table 5 shows the mean performance parameters obtained in

different experiments. Based on the performance the proposed work and their comparative algorithms the proposed technique found efficient and accurate. Additionally, it produces accurate results with less resource consumption. This model is further extended in the near future with the following extensions:

1. The proposed system is implemented with the crop plant disease prediction and recommendation system
2. The proposed model includes the disease system for suggesting the appropriate solution discovery and recovery

The proposed work further also includes the optimization techniques for optimizing the solutions with respect to the problem attributes.

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Table 1 observation matrix

	Cluster 1…	Cluster N
State 1	$Values_{11}$	$Values_{N1}$
State N	$Values_{1N}$	$Values_{NN}$

Table 2 transition matrix

	State 1	State N
State 1	$Probability_{11}$	$Probability_{N1}$
State N	$Probability_{1N}$	$Probability_{NN}$

Table 3 proposed algorithm

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Table 4. shows the performance of the

PM = Proposed Model, AA = Apriori algorithm, CM = Collaborative methods											
	Accuracy			Error Rate			Memory Usage			Training Time		
Data set size	PM	AA	CM	PM	AA	CM	PM	AA	CM	PM	AA	CM
100	82.6	74.1	75.4	17.4	25.9	24.6	13882	14048	13968	78	89	83
200	84.2	77.9	76.2	15.8	22.1	23.8	14006	14382	14257	124	145	133
300	83.7	76.3	78.5	16.3	23.7	21.5	14325	14671	14428	161	189	177
500	86.1	74.8	79.9	13.9	25.2	20.1	14419	14884	14764	280	335	310
700	89.4	78.4	80.1	10.6	21.6	19.9	14646	15363	14977	387	469	421
1000	93.9	79.1	82.5	6.1	20.9	17.5	14819	15366	15225	503	733	653
1500	96.2	80.5	84.7	3.8	19.5	15.3	14904	15862	15563	728	987	864

Table 5. performance summary

Parameters	Proposed model	Apriori algorithm	Collaborative methods
Accuracy	88.01 %	77.3 %	79.61 %
Error rate	11.98 %	22.7 %	20.38 %
Memory usages	14428.71 KB	14939.42 KB	14740.28 KB
Time usages	323 MS	421 MS	377.28 MS
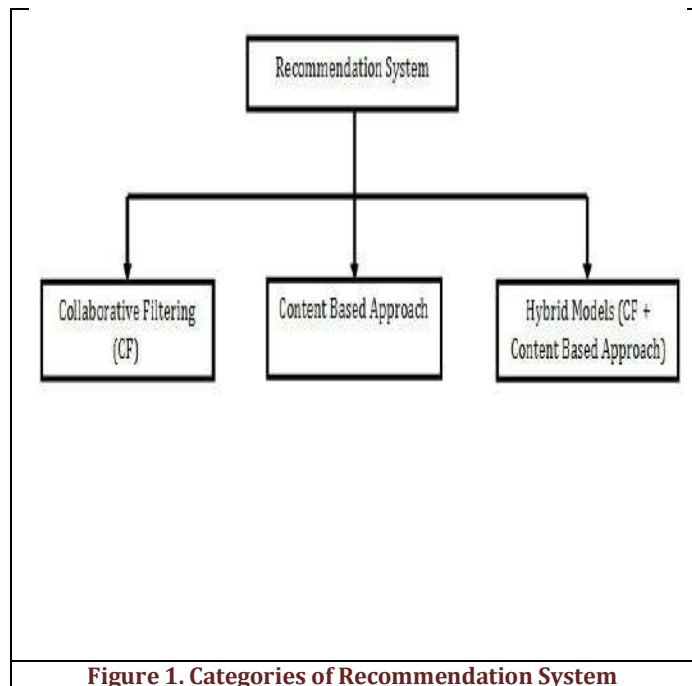
Figure 1. Categories of Recommendation System   **Figure 2. proposed model**
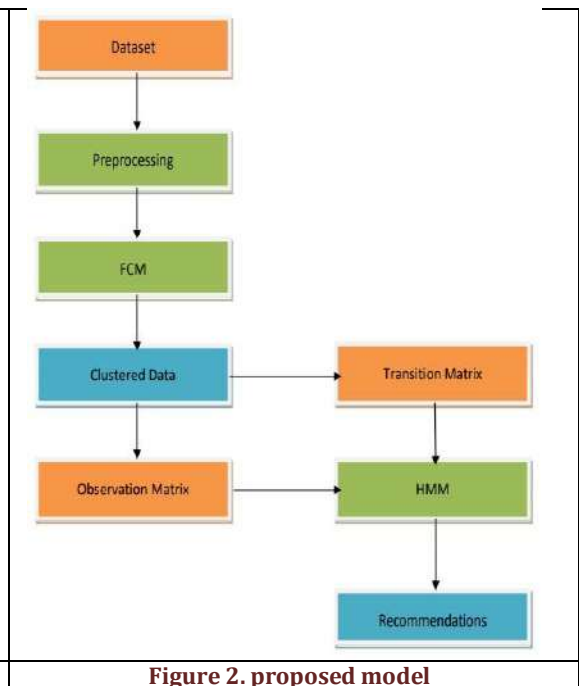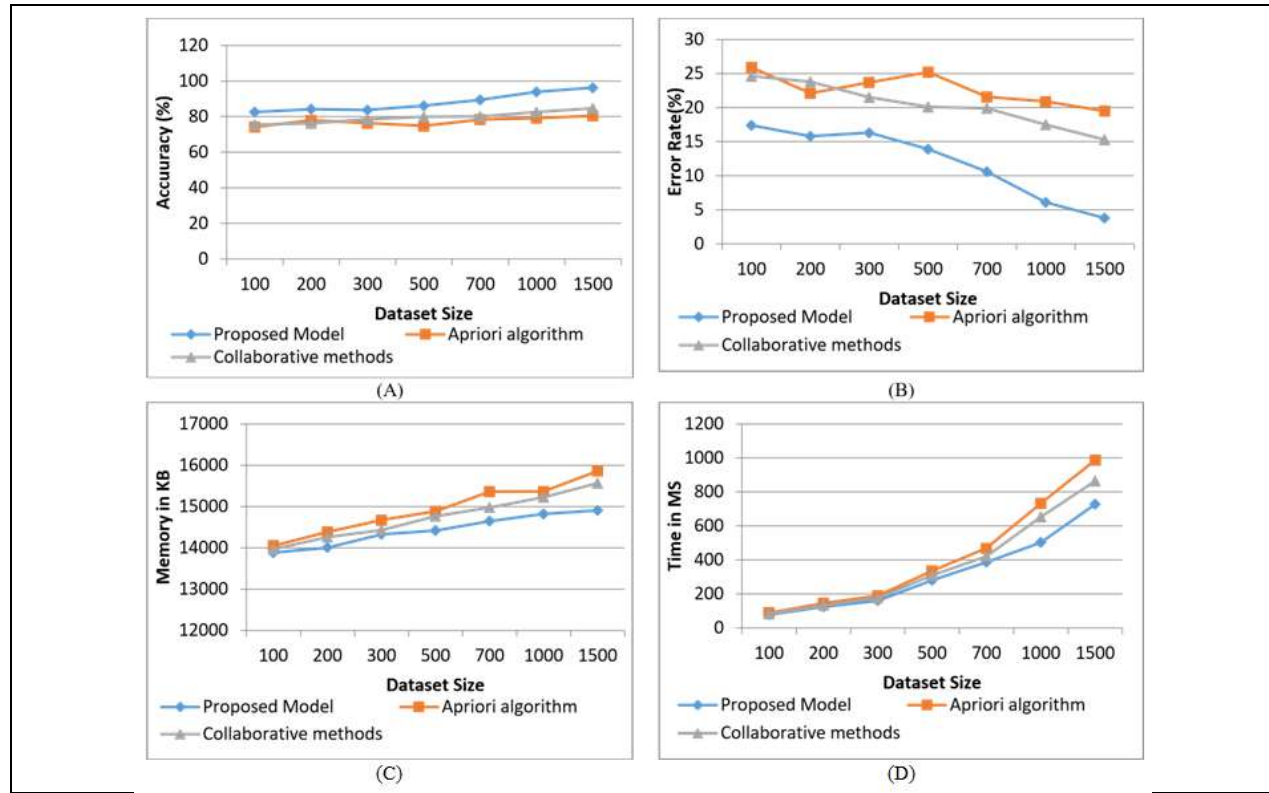
Figure 3. Provides the experimental results of recommendation models with the help of (A) Accuracy (B) Error Rate (C) Memory Usage (D) Time consumption

REVIEW ARTICLE

Effect of Varma Points Manipulation and Oleation Therapy for Uthiravatha suronitham –A Case Study

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ABSTRACT

Uthira Vatha suronitham is one of the vathadisease in Siddha medicine. The clinical features as per Siddha are swellings of joints, unable to walk, pain and swellings in small phalanges. These signs and symptoms can possibly compare to Rheumatoid arthritis (RA) which is a auto immune disorder mainly affect small joints of the hands and feet in a symmetrical pattern.Varma is one of the mode of therapy of physical manipulation either by applying pressure on the varma points or using with specific medicated oils. Varmam is the electro-thermal points where life energy (vaasi) flows throughout the body via particular pathway which is concentrated on two bones joint or a muscle inserts into a bone or the blood vessels and nerves are prominent. This study aims to explore the effectiveness of Varma physical manipulation therapy along with oil application for Uthira Vathasuronitham with special reference to the rheumatoid arthritis. A 45 years old female suffering from pain ,stiffness and swelling in right and left elbow, knee joints and hands for 3 months came to arthritis clinic at Agasthiyar hospital, Trincomalee. Classification of grading of assessment criteria was adopted to diagnose as Rhematoid artiritis. Varma therapy was carried out every day for one month. Assessment   was carried out on the basis of sign and symptoms and laboratory investigation. The pain was gradually reduced during the treatment as well as other symptoms like morning stiffness, swelling, heat, restricted movements also well responded after one month of treatment. After Varma therapy , the Narayana oil application was continued for further one month. During the course of treatment and one month follow up period diet and regimen were followed strictly. It is concluded that the symptoms were significantly reduced to the varma physical manipulation and it was controlled with application of Narayana oil.

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Key words: Rheumatoid Arthritis, Life energy, pain, swelling, Electro-thermal points

INTRODUCTION

Uthira Vathasuronitham is one among the 80 types of vatha diseases, as per siddha reputed text the clinical features are swellings of joints, unable to walk, pain and swellings in small phalanges, excessive salivation, gradual increase of swellings and drowsiness [1]. Among 32 external therapy, the varma is one of the treatment mode in Siddha system of medicine which is popular in south India and Sri lanka at nearly10,000 years old. This therapy is involved with physical manipulation either by applying pressure on the varma points and/or using massage with specific medicated oils. Life energy (vaasi) flows throughout the body via particular pathway. This is concentrated where two bones join or a muscle inserts into a bone or the blood vessels and nerves are prominent which is called electro-thermal points. Varma points are manipulated over the superficial nerves, blood vessels, bony prominent and muscular tissues [2]. Ancients siddhars knows these vital points by their spiritual knowledge and manipulate these points to cure many illnesses. But these points when hit forcefully produce injurious effect called Varma injuries. Varma therapy is most effective in neurological disease , endocrine diseases, metabolic diseases and diseases of locomotor system.Varma energy pathway is known as electro-thermal conduction which can be correlated scientifically with physiological analgesic pathway (brain opiate system). Marta Imamura et al mentioned in her research findings the massage or pressure therapy will stimulate to release neuro modulators such as Endrophins, Encephalins and Dynorphins [3].

Rheumatoid arthritis (RA) is the most common persistent inflammatory arthritis and occurs throughout the world and in all ethnic groups which affects 0.5 – 3 % of the population and common age group is 30-50 years. [4]. RA has a predilection to affect women, in whom incidence and prevalence rates of RA are twice as high as in men.The lifetime risk of developing RA is 3.6 percent in women and 1.7 percent in men. It is an immunological disorder of connective tissue triggered by environmental factors, in patients with genetic predisposition. Disease course is variable with exacerbations and remissions. It is a chronic, systemic, autoimmune inflammatory condition causing synovitis and resulting in pain, swelling, warmness, tenderness and stiffness. Pain and stiffness often worsen after rest [5]. Most commonly wrist and hands are involved with symmetrical nature, where joints lined by synovial membrane may be involved initially and extra articular involvement of organs as skin heart, lungs and eyes can be significant. In the joint, synovial capsule became inflamed, causing damage to the joint tissue. These tissue damage can cause chronic pain and deformities. It is a chronic systemic inflammatory polyarthritis that primarily affects small joints of the hands and feet in a symmetrical pattern [6].

Morning joint stiffness (MJS), may follow a circadian pattern and the rhythm of pro-inflammatory cytokines, such as interleukin-6 (IL-6).The increase in nocturnal anti-inflammatory cortisol seen in patients with RA is generally insufficient to suppress the ongoing joint inflammation, often resulting in joint stiffness in the morning. Morning joint stiffness is included in the original ACR classification of RA and remission criteria.[7] A musculoskeletal exam nearly exclusively relies on history, inspection and palpation of the joints and some specialized tests involving those techniques. The history and physical examination are valuable tool to arriving at an accurate diagnosis of musculoskeletal disorders. In the diagnosis of musculoskeletal disease, 60% of the weight might be placed on the history ,30% on the physical and 10% on laboratory data. The key features to note and record on the examination of the joints are Pain, Morning joint stiffness, Swelling, Tenderness and Loss of motion. Other important physical signs are temperature and color changes over the joint.[8] Stiffness was reported to be a normal part of having RA, experienced in joints and more widespread, related to behavioural and environmental factors and tohave marked variability which is not being limited to early morning. It resulted in wide-ranging consequences that had a major impact on patient's daily lives and necessitated self-management. Patients placed greater importance on stiffness

impact than stiffness severity or duration. Stiffness is internationally recognized as an important indicator of inflammatory activity in RA.[9]

METHODOLOGY

Case report
45 years female from Trincomalee town, management assistant attended to the arthritis clinic on 02.03.2020.

Present complaints
Chief complaints of pain and swelling in both elbow and knee joints and only pain in both hands for 3 months.

Present History of Illness
Pain and stiffness marked in early morning for more than 2 hours. The pain increases even in routine activities and she was disturbed due to chronic intermittent pain. Pain is aggravated by hard work, walking long distance, climbing up stair cases , cold climate, cold bath and intake of gaseous meals, unable to perform routine daily works for last 2½ months .

Scope of Clinical examination
Swelling is present in right elbow, right knee joint. On palpation tenderness and heat are positive in right elbow joint and right knee joints. Mild heat is present in left elbow joint. Pain and swelling and stiffness in the third and fourth finger of right hand. Movement of the joint is restricted in right elbow joint and right knee joints. These symptoms are assessed by VAS pain score and likert scale. Examination of cardiovascular system, respiratory system , gastro intestinal system , endocrine system and uro- genital systems are normal .

Past history
Same complaints occurred within 5 years period with frequent remissions and flares. Initially pain developed in fingers and gradually developed pain and swelling in fingers, both elbows and knee joints. Later stiffness, loss of movement, pallor and tiredness also developed. There is no history of diabetes mellitus, Tuberculosis, bronchial asthma and trauma. Patient followed the allopathic medicine, using NSAIDs and corticosteroids for last 3 years. But the codition was not reduced completely and on and off type of pain was developed.

General Examinations:
Patient looks normal, Pulse: 80 / minutes, Blood pressure:110/ 80 mm Hg,Temperature:38.2° C Respiratory rate:20 beats / minutes, Pedal edema:nil, Clubbing finger: nil, Pallor: mild,Cyanosis: nil, Jaundice - nil

Diagnostic scope
The provisional diagnosis was made as Rheumatoid arthritis of joints using the following criteria. The American College of Rhematological classification criteria for RA of the knee synovitis (swelling) with at least one of the following three items:[10]
- Age 30-50 years
- Morning stiffness ≥ 1hour duration
- Minimum 3 joints involvements both large and small
- RA score 6/10

Classification of grading of assessment criteria was adopted to diagnose as Rhematoid artiritis.(having 10/10 score as per criteria for RA.ACR,2010)(Table 1).[11]

Scale assessment for Signs and symptoms

Rating of signs and symptoms such as swelling, tenderness and loss of motion can be graded conveniently on a scale of 0-4. In general terms, 0-normal, 1-mild abnormality, 2- moderate, 3- marked and 4-maximum abnormality. Pain assessment tool is intended to help patient care providers assess pain according to individual behavioural observation or face expression to interpret and expressed pain intensity.0-10 VAS Numerical scale is used.[12]

VAS Numerical Pain Scale

Therapeutic scope

- Preparatory stage - Purgation therapy for one day *Agastiyar kuzhambu* 65mg with warm water was given as purgation therapy in preparatory stage which helps to cleansing the body.

Treatment Intervention

- *Varma therapy* – Pointed digital pressure on specific points for one month, two times a day
- *Narayana* oil application – External oil application for affected area for two times for a day. Treatment intervention was commenced with varmatherapy and later *Narayana* oil application.

Varma therapy

Varma therapy was given to the patients every day for one month. Weekly improvements of signs and symptoms were recorded.

Oleation therapy

Narayana oil was applied gently for the affected area two times a day for further one month followed by varma therapy. Weekly improvements of signs and symptoms were recorded. Diet pattern and life style modification were advised. The follow up period was one month while no remissions and exacerbations was observed. Following Varma points were manipulated for Varma therapy.

RESULTS AND DISCUSSION

The grading scale presented here enables the assessment of efficacy of combined varma and oil massage therapy. A 45 years old female residing at town area and she was suffering from signs and symptoms of rheumatoid arthritis such as joint pain, difficulty to move the particular point of the joints with morning stiffness over MCP (metacarpo-phalangeal joint), wrist joints, elbow joints, knee joints and also disturbed sleep due to pain at night. She was unable to move without support in early morning due to severe pain and morning stiffness for more than 2 hours. Pain and mild swelling started from bilateral wrist joints 5 years before. Later on pain started to radiate to other joints. The clinical examination and x-ray of joints confirmed the diagnosis as rheumatoid arthritis. Agasthiyar kuzhamabu 65mg with warm water given for purgative therapy for cleansing the bowel. Thereafter varma therapy was started and continued for I month. The improvement was noticed. How ever 2nd month of treatment Narayana oil was applied for affected area. The signs and symptoms were reduced further. The 3rd month was observation period while symptoms, completely reduced and no remission occurred.

Outcome = Post treatment score – Pre treatment score

The cardinal signs and symptoms of RA such as pain , swelling, Morning joint stiffness(MJS), warmness and loss of movement in the both right and left elbow, and both right and left knee joints were assessed by standard parameters. The pain score(Table 2) evaluated by VAS numeric pain scale was reduce from very severe pain(P_9) in Right elbow and left elbow severe pain(P_6) which were reduced into no pain (P_0),reduce from severe pain(P_6) in Right knee and left knee severe pain(P_6) into no pain (P_0),reduce from moderate pain(P_4) in Right MCP and left MCP moderate pain(P_5) into no pain (P_0). Reduce from moderate pain (P_5) in Right PIP and left PIP moderate pain (P_4) into no pain (P_0). The tenderness is evaluated by palpation.(Table 3)Severe pain on mild palpation (T_2) in both

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right and left elbow but after treatment it become pain free on palpation. (T_0). Right and left knee shows severe pain on mild palpation(T_2)but after treatment tenderness was relieved. (T_0).More severe pain on mild pressure (T_3)in right MCP and severe pain on mild palpation in left MCP. (T_2). Both side tenderness were relieved after treatment. More severe pain on mild pressure (T_3) in right PIP and severe pain on mild palpation in left PIP. (T_2)Both side tenderness were relieved a

Swelling was obvious on casual observation in right and left elbow (S_2) but both were reduced after treatment (S_0). Markedly abnormal swelling was noticed in right knee (S_3) and left knee swelling was obvious on casual observation (S_2) but both were relieved after treatment. Swelling may not be apparent on causal inspection but recognizable to an examination in right MCP (S_1) it was obvious on casual observation in left MCP (TS_2). In Right and left PIP Swelling may not be apparent on causal inspection but recognizable to an examination (S_1) but both were relieved after treatment (Table 4).

Morning joint stiffness lasting ≤ 120min(JS_3) in right elbow and lasting ≤ 60min(JS_2) in left elbow were reduced to normal(JS_0).lasting ≤ 60min(JS_2) in right knee and lasting ≤ 60min(JS_2) left knee which was reduced to normal.(JS_0)In right MCP lasting ≤ 120min(JS_3) Left MCP lasting ≤ 60min(JS_2) which was reduced to normal.(JS_0)In right PIP lasting ≤ 120min(JS_3) Left PIP lasting ≤ 60min (JS_2) which was reduced to normal.(JS_0).(Table 5). Thermal sensation of right elbow was hot (TS3) and left was warmth (TS2) but both were relieved after treatment. Similarly, right knee was warm (TS2) and left knee was slightly warmth(TS1) but both were relieved after treatment. Right MCP was warmth (TS2) and left knee was also warmth (TS2) but both were relieved after treatment. Right PIP was slightly warmth (TS1) and left PIP was warmth (TS2) but both were relieved after treatment (Table 6). A small amplitude movement performed at the limit of the range in right elbow (R_3) and left elbow was not restricted movement (R_2) but movement of both elbow were increased after treatment. In right and left knee were not restricted (R_2) but movement of both knee were increased after treatment. A large amplitude movement performed up to the limits of the range (R1) was noticed in right and left MCP similar in right and left PIP which were increased in to normal.(Table 7)

After the completion of varma therapy for 30 days patient was allowed to take rest for two weeks while she was under observation for symptomatic monitoring. The complaint after 10 days the slight pain was noticed in right elbow and right knee joints. On examination tenderness and slight movement restriction observed. Then patient was asked to apply Narayana oil at affected area for two times a day for one month. After completion of oleation therapy 90% relief on pain, morning stiffness and swelling. Assessment was done after 1 month oil application. After two months of resting period of completion of treatment. Same evaluation protocol was followed to find out the remission of signs of treatment. On follow-up after Six months of second session patient felt significant relief in the sign and symptoms pain, stiffness was markedly reduced and patient felt 100% releif. There is no exacerbations and remissions and Patient feels healthy. Rheumatoid factor and CRP were positive before treatment. But after treatment Rheumatoid factor was negative, no change in CRP. ESR was 44 mm / hour before treatment and it was reduced as 4 mm/ hour after treatment.

CONCLUSION

The Varma therapy plays the role in significant reduction of very prominent signs and symptoms of Rhumatoid Arthritis (Uthira vathasuronitham) such as pain, swelling, tenderness, warmthness, morning stiffness and joint movement but after the oleation therapy complete relief was noticed. Hence it is concluded combined varma point manipulation and oleation therapy were more effective than single varma therapy for Rhematoid arthiritis.

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Table:1 Criteria for diagnosis of rheumatoid arthritis

	Standard Criteria	Standard Scores	Patient's score
1.	**Affected joints**		
	1 large joint	0	-
	2-10 large joints	1	1
	1-3 small joints	2	-
	4-10 small joints	5	5
2.	**Serological Test**		
	Negative RF and ACPA	0	-

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	Low positive RF or ACPA	2	2
	High positive RF or ACPA	3	-
3.	**Duration of symptoms**		
	Less than 6 weeks	**0**	**-**
	Greater than 6 weeks	**1**	1
4.	**Acute phase reactants**		
	Normal CRP and ESR	0	-
	Abnormal CRP or ESR	1	1
	Total score		**10**

Table:2 Score value of pain(VASNumeric pain scale)

Pain grading	Grade
No pain	P_0
mild	P_{1-3}
moderate	P_{4-5}
severe	P_6
Very severe	P_{7-9}
Worst pain	P_{10}

Table:3 Score value ofJoint Tenderness[13]

Tenderness	Grade
No pain on pressure	T_0
Slight or mild tolerable discomfort on palpation	T_1
severe pain on mild palpation	T_2
More severe pain on mild pressure	T_3
Intolerable pain even on mild touching caused by spontaneous touch stimulus	T_4

Table:4 Score value ofJoint swelling[14]

Swelling	Grade
No swelling	S_0
Swelling may not be apparent on causal inspection but recognizable to an experienced examiner	S_1
Swelling obvious on casual observation	S_2
Markedly abnormal swelling	S_3
Swelling to a maximally abnormal	S_4

Table:5 Score value Morning Joint stiffness (MJS) [15]

Duration of MJS	Grade
No stiffness	JS_0
Lasting ≤30min	JS_1
Lasting ≤60min	JS_2
Lasting ≤120min	JS_3

Table:6 Score value for warmthness [16]

Thermal sensation	score
Cold	TS_{-3}
Cool	TS_{-2}
Slightly cool	TS_{-1}
Neutral	TS_0
Slightly warmth	TS_1
Warmth	TS_2
Hot	TS_3

Table: 7   Score value Joint movement - Maitland's 5 Point Scale [17]

Range of Joint movement(RJM)	Grade
a small amplitude movement at the beginning of the movement	R_0
a large amplitude movement performed within the free range but not moving into any resistance (perceived stiffness)	R_1
a large amplitude movement performed up to the limits of the range	R_2
a small amplitude movement performed at the limit of the range	R_3
a small amplitude movement performed beyond the limit of the range (a manipulation)	R_4

Table:8 Varma points in elbow joints and MCP & PIP Joints: .[18]

	Varma points	Location
1.	Savvuvarmam	four finger breath distal from shoulder joint on the medial side of upper arm
2.	Sudotharivarmam	four finger breath above the Manibanthavarmam (radial aspect of the forearm)
3.	Manibanthavarmam	Middle of the wrist (ventral aspect)
4.	Ullankaivellaivarmam	At the junction of big and second toe .
5.	Thatsanaikkalam	Below the middle finger (ventral aspect)
6.	Kavulikkalam	web area between the thumb and the index finger .

Table 9:Varma points in knee joints.[19]

	Varma points	Location
1.	Ullangaalvellaivarmam	At thejunction of big & 2nd toe in the planta region
2.	Viruththikkalam	Situated between big toe and adjacent in its dorsal aspect.
3.	Padankaalvarmam	one finger breath bellow lateral malleolus
5.	Uppukkutryvarmam	Situated 3 finger breaths above posterior aspect of the heel.
6.	Kuthikkalvarmam	Situated 7 finger breaths above posterior aspect of the heel
7.	Mudduvarmam	Situated in centre of the popliteal fossa.
8.	Panchamugavarmam	5 points – around the patella
9.	komberikkalam	8 finger breath above the medial malleolus

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Table 9 Physical findings:

Joints	Pre Treatment	Post Treatment	Pre Treatment	Post Treatment	outcome
Side	**Right**	**Right**	**Left**	**Left**	
Elbow	P_9 S_2 T_2 JS_3 TS_3, R_3	P_0 S_0 T_0 JS_0 TS_0, R_0	P_6 S_2 T_2 JS_2 TS_2, R_2	$P_0, S_0, T_0, JS_0, TS_0, R_0$	Marked improvement
Knee	P_6 S_3 T_2 JS_2 TS_2, R_2	P_0 S_0 T_0 JS_0 TS_0, R_0	P_5 S_2 T_2 JS_2 TS_1, R_2	P_0 S_0 T_0 JS_0 TS_0, R_0	Marked improvement
MCP	P_4 S_1 T_3 JS_3 TS_2, R_1	P_0 S_0 T_0 JS_0 TS_0, R_0	P_5 S_2 T_2 JS_2 TS_2, R_1	$P_0, S_0, T_0, JS_0, TS_0, R_0$	Marked improvement
PIP	P_5, S_1, T_3 JS_3, TS_1, R_0	P_0, S_0, T_0 JS_0, TS_0, R_0	$P_4, S_1, T_2, JS_2, TS_1, R_0$	$P_0, S_0, T_0, JS_0 TS_0 S_0, R_0$	Marked improvement
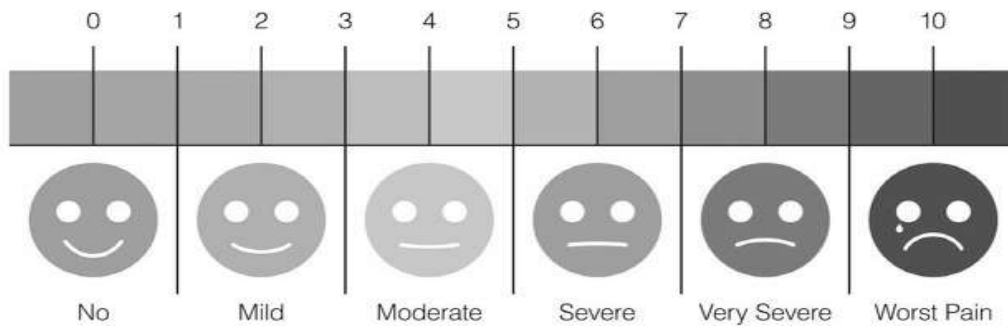
Fig. 1. VAS Numerical Pain Scale

RESEARCH ARTICLE

Internet of Things for Future Technology Enhancement

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ABSTRACT

Mobile devices are common these days, and over the next few years, online objects will expand in every field. Producers have implemented the Internet of Things in savvy domestic gadgets, cars, cities, and industrial facilities, and specialists foresee their utilization will proceed to develop the internet of things. Internet of things has made it through its rising a long time where organizations needed to be a portion of the buildup but didn't fundamentally know what to do with the innovation. These times organizations and businesses took activity with particular targets to create more compelling supporting innovation. Indeed organizations can spend less on fetched since they have numerous alternatives, like Wi-Fi, low-power. More prominent implanted preparing control fits into indeed littler gadgets and information preparation can take all through the information pipeline from the sensors to the cloud. Coordination administration systems and stages have given organizations the capacity to analyze expansive amounts of information.

Keywords: Internet of Things, Wi-Fi, Cloud computing, Big Data.

INTRODUCTION

"Internet of Things" (IoT) is the interconnected network of physical objects with embedded electronic systems for interacting with each other and sensing the external environment through communication and sensing interactions within their architecture. The Internet of Things is going to revolutionize the way people lead their lives during the next few years by delivering advanced services and providing real-world applications. A few of the many categorical examples where IoT has been successful are in the fields of medicine, power, gene therapy, agriculture, smart cities. The Internet is connected to over 9 billion Things (physical objects) at present. The number of mobile devices will rise to 20 billion in the near future [1].IoT is a method of extending the internet to more dumb devices, such as toasters, that are currently not connected to the internet. A person with a disability monitor implant, an animal with tracking devices, or anything else that can be connected to an internet connection is considered "smart" by way of IoT [2]. Extensive research has been going on and done in scientific journals and press reports on both the potential

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effectiveness and application of IoT transformations. It could be used as preliminary implementation before making innovative business plans while considering Security, Assurance, and good coaction. The IoT time period was introduced in 1999 with the help of Kevin Ashton. The term describes a number of layers used in IoT, as well as a few related terms. The IoT environment, for example, can identify our family's regular gadgets as a Smart-Home when our regular family gadgets enter the net. It is not merely a future science fiction vision. It is a reality now. In addition to pure technical development, this has a wider impact [3].

ARCHITECTURE OF IoT
In IoT, data is gathered and transmitted over the internet by devices that connect to it. IoT systems connect highly specialized devices with a limited degree of programmability and customization in comparison with conventional cyber systems that connect computers for general purposes. Instead of centralized storage and processing resources in huge Data centers, IoT systems generally store and process information in a scattered way. Unlike cyber-systems, Internet of Things systems use sensors and actuators to collect data from the physical world. These are referred to as cyber-physical systems. From the security and privacy standpoints, IoT creates new opportunities and risks. To date, industry and consumers have only just begun to advance technology. IoT is a vast universe of strange devices. Each physical unit is equipped with sensors, which continuously transmit information about how the unit is operating. In IoT, all these devices or sensors are provided with a platform for dumping their data into a common communication language between them. Sensors and communication services (gateways, modems, and routers) are critical elements of IoT hardware as are battery power and touch screen power. There is no widely accepted architecture for the IoT ecosystem as each company has its own set of criteria [3].

The further part of this article contains "Literature survey" which will give state of the art overview on important studies that highlights various drawbacks and problems in IoT; followed by "IoT architecture and technologies" where in the IoT functional block and architecture is discussed. Then "Major Key issues and challenges of IoT" is discussed; followed by "Major IoT applications" which provides emerging application domains of IoT. Next "Importance of big data analytics in IoT" is discussed where, the role and importance of big data and its analysis are discussed.

Literature survey
The Internet of Things has a multidisciplinary view to provide its benefits to a variety of areas, such as the Environmental, industrial, medical, public private sectors. Various research scientists have discussed IoT technology based on their interests and concerns. In Figure 2, it is illustrated that IoT has many potential applications. Its power and potential can be seen in several different applications domains. There have been many important projects in the IoT sphere in recent years. Some of the most innovative IoT projects have acquired the market share can be seen in Fig. 3. In Fig. 3 American, European, and Asia/Pacific regions are the top three locations for IoT projects. There is a correlation between contributions of the America to health care and supply chain projects, and the contribution from the European countries towards smart city projects [4]. IoT market share worldwide is shown in figure 4 [4]. IoT-based projects in the energy, smart city, smart vehicle, and smart city sectors all hold significant market shares compared to other sectors

Smart city applications include smart homes as well, making them one of the trending application areas for IoT. In smart homes, there is IoT embedded appliances, air conditioner and heating appliances, TVs, streaming devices, and security systems that are communicating with each other to provide the highest level of comfort, Assurance, Security, and reduced energy consumption. Through a central control unit based on IoT and connected to the Internet, all communication takes place. Smart city is an emerging concept that has been the subject of much research activity in the last decade. There will be a 100 billion dollar smart home business economy by 2022 [5]. Smart homes are comfortable and also provide the house owner with substantial cost savings in many ways including comparatively lower electricity bills due to low energy consumption. Aside from smart homes, smart vehicles also fall under the category of a smart city. Latest Generation cars which are equipped with smart devices and sensors which controls various components like steering, headlights and other parts of car to the engine [6]. Extensive

research in IoT sector is committed in the development of replacement smart car systems that come with wireless communication between vehicle-to-vehicle and vehicle-to-driver to make sure conditional monitoring with a safe and comfortable driving experience [7]. Khajenasiri et al. [5] conducted a survey on the IoT solution for smart energy control to help smart city applications. They conveyed that the Internet of Things has been implemented in only a couple of implemented areas to help the technology and people. The scope of Internet of Things technology is very vast in future; IoT is in a position to conquer most application area. They revealed that reduced energy consumption is one of the important aspects of society. IoT can help in the development of a smart energy system that can save energy and money. As part of the smart city concept, they presented an IoT system architecture. Often IoT hardware and software are immature, so this can pose a challenge to implementing this. In order to achieve stable, coherent, and user-friendly IoT systems, these issues must be solved.

Alavi et al. [8] presented the development of urban area issues in the cities. Growing urban populations resulted from people moving from rural to urban areas. Consequently, smart solutions are needed for mobility, energy, healthcare, and infrastructure. IoT developers are interested in creating smart cities, which is one of their most important applications. In this report, various topics are examined including traffic management, air quality management, road safety solutions, smart parking, smart lighting, and intelligent waste management (Fig. 5). Researchers mentioned that the Internet of Things is tackling these challenges head-on. Smart city technologies have opened doors for entrepreneurs in the field due to the growing need for improved infrastructure for smart cities. For the development of sustainable smart cities, IoT-enabled technology is essential. Authenticity and privacy of user info is also a key issue of IoT that requires extensive measures in research. Weber [9] stressed the importance of incorporation of data security, access control. The added benefit of ensuring confidentiality of client information, as well as resilience to attacks, could be an added benefit. There are other concerns that need solutions to strengthen safety in IoT, such as authentication and access control. Authentication is of great importance to prevent the loss of confidential information. Liu et al. [11] discussed a way to handle authenticating and controlling access. An authentication scheme has been proposed by Liu et al. that supports Elliptic Curve Cryptosystem and is tested against various eavesdropping threats, man-in-the-middle attacks, key control attacks, and replay attacks.

As a result of their research, they claimed their proposed schemes are capable of enhancing authentication and access control in IoT communications. Meanwhile, Kothmayr et al. proposed a two-way authentication scheme for IoT communications based on datagram transport layer security. A web-based attacker is constantly looking for ways to steal the secured information. An IoT based communication network is designed to offer message security, integrity, authenticity and confidentiality, low overhead, and end-to-end latency. An approach proposed by Li et al. [13] for data-centric IoT applications based on cloud platforms was proposed. IoT applications that run on cloud platforms require efficient solutions for an appropriate device, software configuration and infrastructure. The IoT community is committed to developing solutions that take into account both large platforms as well as the heterogeneous nature of IoT objects and devices. Olivier et al. [14] demonstrated how a SDN-based architecture is more secure and flexible for the Internet of Things, although a clearly defined architecture is not readily available. The authors proposed that SDN-based security architecture is more efficient and flexible.

According to Luke et al. [15], the most important tasks of a secure sensor network (SSN) are to protect data from replay attacks and to provide authentication Their study discussed TinySec [16] and ZigBee [17]. Both are efficient and reliable SSN services, but ZigBee is backed by a range of security certifications that have been certified by the U.S. Department of Homeland Security. In comparison, J Big Data (2019) offers higher security but consumes much more energy, while TinySec consumes much less energy but is less secure than ZigBee. Yan et al. [18] state trust management is an important issue in IoT and propose another architecture MiniSec that supports high security and low energy consumption. In an investigation into trust management, they discussed its importance as it pertains to developers and users of IoT services and applications [19]. Noura et al. [20] stated the importance of interoperability in IoT because it allows the integration of devices, services from different heterogeneous platforms to supply efficient and reliable service. Several other studies focused on the importance of interoperability and discussed several challenges that interoperability issue is facing in IoT [21, 22, 23].

Kim et al. [24] proposed an IoT-based ecological monitoring system to address the problem of global climate change. They noted that current approaches are difficult and require a lot of human effort. It is also necessary to conduct routine visits to the location under investigation so that the sensors installed there can supply useful information. Additionally, some information is missing which results in our findings not being highly accurate. A network based on IoT may thus be able to solve this problem and provide highly accurate analysis and predictions. Later, Wang et al. [25] discuss issues of domestic wastewater treatment. IoT is often very effective in the treatment and monitoring of wastewater and the process of monitoring wastewater. They discussed several deficiencies in the treatment and monitoring process and proposed solutions that supported IoT. It has several important aspects including geographical, ecological, and social. Agriculture is one of the most important domains in the world.

IoT architecture and technologies
The structure of the IoT system has 5 different layers that define multiple functionality. These layers are the vision layer, the network layer, the middleware layer, the application layer, the business layer. Beneath the rock of IoT architecture, the vision layer contains sensory data and business rules. the information collected by these devices is provided by a network layer, which serves as a transmission point for conveying information from the point of view to the information processing system. The next level layer referred to as middleware is responsible for data transfer using any wireless or wireless method, including 3G/4G, Wi-Fi, Bluetooth, etc. supports computer-generated results everywhere, this layer processes information obtained from the network framework to make decisions. This information is then used in the global device management framework. At the top of the architecture, there is a business platform that controls the IoT system, its use and services. The business base visualizes the information and statistics obtained from the electronic component and uses this information to plan the next phase of the process and methods of supply. In addition, IoT structures are often modified according to the need and application environment [14, 25, 32]. In addition to the horizontal framework, the IoT system also contains a number of functional blocks that support a variety of IoT functions such as hearing, verification, detection, and management and control [33]. Fig. 6 shows such functional blocks of IoT architecture. Each of these functional blocks contributes to efficient IoT systems such as I / O functionality, communication, processing, audio / video monitoring, and storage management. Collectively, these functional blocks form a highly efficient IoT system that is essential for improving efficiency.

Although, there are a few reference and proposed technological structures, but these are still far from the standard IoT standard for global [34]. Therefore, a suitable structure still needs a design that can meet the global IoT requirements. The typical operating structure of the IoT system is shown in Fig. 7 which shows the dependence of IoT on the parameters of a particular system. IoT portals play an important role in IoT connectivity because it allows communication between IoT servers and IoT devices associated with several applications [35]. In an IoT architecture, it is necessary to address several constitution problems such as scalability, modularity, and collaboration. In order to form the IoT structures, the users will need direct and uncomplicated capabilities with easy-to-use applications, as well as domain interaction with multiple systems. Furthermore, the architecture must be able to increase performance and integrate some automation with IoT devices within the system. Moreover, the increase of data generated by IoT devices could pose a new challenge due to the increasing amount of big data available. IoT systems have large amounts of data to consider, so effective architecture is imperative. Fig. 8 shows an illustration of how the modern IoT technology system architecture can be characterized as four-stage architecture. Cloud and fog technology work together to support the management, monitoring, and analysis of large amounts of data in IoT systems.

Fig.6 illustrates such functional blocks of the IoT architecture. Each of these functional blocks contributes to efficient IoT systems like I/O operation, connectivity, processing, audio/video monitoring, and storage management. Taken together, these functional blocks form a highly efficient IoT system critical to optimizing performance.

Major key issues and challenges of IoT
The involvement of IoT based systems altogether aspects of human lives and various technologies involved in data transfer between embedded devices made it complex and gave rise to many issues and challenges. These issues also

are a challenge for the IoT developers within the advanced smart tech society. As technology is growing, challenges and wish for advanced IoT system is additionally growing. Therefore, IoT developers got to consider new issues arising and will provide solutions for them.

Security and privacy issues

Due to several threats, cyber-attacks, risks, and vulnerabilities, security and privacy is amongst one of the most important concerns within the IoT. Insufficient authorization and authentication, insecure software, firmware and web interfaces are the problems that compromise device level privacy [37]. With regard to various aspects of IoT Systems, security and privacy are crucial parameters for developing confidence [38]. Including security mechanisms at every layer is essential to stopping security threats and attacks in IoT architecture. In order to ensure the safety and privacy of IoT based systems, several protocols are developed and deployed on every layer of the channel. With Datagram Transport Layer Security (DTLS) and Secure Socket Layer Security (SSL), IoT security solutions can be delivered at the transport and application layers [39]. Wireless communication within an IoT system can also pose security risks if it utilizes wireless technologies. However, some IoT applications require different communication methods to ensure the safety of devices communicating. There are therefore requirements for deploying certain mechanisms for detecting, self-restoring, and recovering malicious actions. On the other hand, privacy is also an issue that allows users to feel comfortable and secure when using IoT solutions. To determine the communication between trusted parties, a secure network is required for authorization and authentication [41]. Another concern is the different privacy policies for objects communicated within the IoT system. Each object should therefore be ready to check whether the privacy policies of other objects in the system are the same before sending the information.

Intercommunication/standard issues

This exchange of data does not depend on the software or hardware used to exchange data. Intercommunication is achieved by the mutual exchange of information and knowledge between various IoT systems. Various solutions used for IoT development create a challenge for the intercommunication issue. In order to enhance the intercommunication of different objects within diverse environments, different functionalities are being provided by IoT systems. The four levels of intercommunication are technical, semantic, syntactic, and organizational [42]. Also, a combination of different IoT platforms with support for their functionality can provide users with various options [43].The intercommunication of systems is an important issue that researchers have addressed with several solutions. Adapters/gateways might be used to implement these solutions, or virtual networks/overlays could be used to implement them, or service-oriented architecture could be used. However, despite the fact that intercommunication handling approaches reduce some of the pressure on IoT systems there remain certain challenges that should be investigated in future [20].

Scalability, availability, and reliability

It is crucial with IoT to manage an outsized number of devices with varying memory, processing power, and storage capacity, so that the system can support new services, equipment, and devices without degrading its performance. Scalability and availability should both be considered within the layered framework of IoT. Both of these are important components of IoT. Adding devices, storage, and processing power as needed to the IoT network through cloud-based platforms is an excellent example of scalability. However, this global distributed IoT network suggests a new research paradigm that would serve global needs through the development of a smooth IoT framework. It is important that authentic objects are always able to access the resources they need regardless of their location and time of need. Small IoT networks are attached to internationally recognized platforms to get access to their resources and services in a distributed fashion. In order to ensure availability, the utilization of various data transmission channels, such as satellite communication, may cause some service interruptions as well as limiting resources. To ensure uninterrupted service and availability of resources, a reliable and independent data channel is required.

Quality of Service (QoS)

In addition to standards, performance, efficiency, and quality of service, the Quality of Service (QoS) and the architecture of IoT devices, systems, and architecture are important factors [29]. Reliability, cost, energy

consumption, security, availability, and repair time are important and required metrics for IoT applications [48]. Moreover, IoT services and devices must be able to meet QoS standards. Moreover, all users can also specify what they need in regard to QoS, thus improving the overall IoT ecosystem. QoS assessment often takes place using several approaches. However, White et al. [49] have pointed out that there is a trade-off between quality factors and methods. As a result, good quality models are necessary to overcome this trade-off. Models like ISO/IEC 25010 [50] and OASIS-WSQM [51] are available in the literature that may be used to evaluate the methods used for QoS assessment. IoT service quality can easily be assessed using these models, as they provide a range of quality factors.

Major IoT Applications
Emerging economy, environmental and health care
Society and other people benefit tremendously from IoT, both financially and publicly. The Internet of Things is making a significant contribution towards the social, economic, and health goals of the United Nations. This includes economic development, water quality maintenance, well-being, industrialization, etc. Sustainable development of the Internet of Things (IoT) is another important issue. IoT developers have to consider the impact of IoT systems and devices on the environment in order to reduce their negative impact [43]. Internet-connected devices are consuming more energy than ever before, which poses a challenge to sustainability. Thanks to internet-connected services and edge-cutting devices, energy consumption is increasing at a rapid rate. In order to make new IoT devices with lower energy consumption rates, this area requires research for the fabrication of high-quality materials. Furthermore, green technologies are used to make energy-efficient devices for the future. IoT devices have proven highly efficient at monitoring several health issues, such as diabetes, obesity, or depression. Researchers are working tirelessly on developing IoT devices that are environmentally friendly and beneficial for human health. There are several studies that take into consideration environmental, energy, and healthcare issues.

Smart city, transport, and vehicles:
With the introduction of smart cities, smart homes, and smart vehicles and transportation systems, the normal civil structure of society is being transformed into a high-tech one. Using supporting technologies such as machine learning and tongue processing, rapid improvements are being made to understand the necessity and use of technology at home. For a smart city to become efficient, several technologies need to be combined with Internet of Things servers, including cloud servers, sensor networks and wireless networks. An important factor is to focus on the environmental aspect of a smart city. Therefore, smart cities should consider energy efficiency and green technologies when planning. Further, the new vehicles feature intelligent devices that can detect traffic congestions and give a driving force a suggestion for an alternative route. Smart devices that are cost-effective should be designed for integration into all ranges of vehicles so that the engine activity can be monitored. Furthermore, it would be beneficial to monitor the activity of the engine in the city. Additionally, the Internet of Things is very effective at maintaining the health of vehicles, and autonomous cars are able to communicate with other autonomous vehicles by using intelligent sensors. As a result, traffic will flow more smoothly than when vehicles are driven by humans in a stop-and-go fashion. However, this process will not be implemented worldwide immediately. The internet of things is expected to contribute to society in the near future with the provision of traffic congestion detection and action. Therefore, a transport manufacturing company should use IoT devices in their manufactured vehicles to become more competitive in the market.

Agriculture and industry automation
Global population is expected to reach a staggering 10 billion by 2050, so agriculture plays an important role in feeding such an enormous population. If agriculture were to play a bigger part in our lives, it could feed an entirely different group of people. As a result, agricultural and technological integration is often required to improve assembly in a more efficient way. Greenhouse technology is an example of how this might be accomplished. In order to enhance production, these technologies take advantage of various environmental parameters. However, manual control of these technologies is a little less effective, requires manual efforts and is more expensive, and results in less production and energy loss. As the Internet of Things advances, smart devices and sensors can be used to monitor and manage the environment inside the chamber, which makes it easier to conserve energy and improve production.

IoT has also enabled industries to automate operations. Industrial IoT includes inventory management, operational control, logistics, supply chain optimization, and chain management.

Importance of big data analytics in IoT

It is estimated that there are over 50 billion connected devices across the Internet of Things. As the network grows and expands, the amount of these devices is growing rapidly. Big data is a term used to describe the massive amount of data transfer between these devices and the internet. The amount of information being transferred over the internet is huge and streaming every second, making it perfect as a big data set. It is inevitable that IoT-based networks will expand continuously, presenting complex issues such as managing and collecting knowledge, storing and processing, and conducting analytics. For smart buildings, IoT big data frameworks provide a significant benefit in tackling several problems, such as managing oxygen levels, monitoring smoke and hazardous gas levels, and adjusting the level of luminosity [54]. By utilizing the sensors installed in buildings, a framework of this type is capable of gathering data and interpreting it to determine the health of the buildings. Further, information analysis and knowledge acquisition techniques are often incorporated into IoT-based cyber-physical systems, which are capable of improving industrial production [55]. Intelligent traffic management systems use IoT devices and sensors to collect and analyze traffic data [56]. When IoT devices and sensors are installed in traffic signals, traffic jams are often analyzed by smart city systems. Health analysis involves analyzing IoT sensors that collect a ton of information about a patient's health condition every second. The large amount of data has to be integrated in one database and processed in real-time so that a quick decision can be made with a high degree of accuracy. This task is best accomplished with the use of large data technologies [57]. The Internet of things in conjunction with big data analytics also has the ability to change the method of doing things in manufacturing industries [58]. Moreover, using cloud computing and analytics in energy conservation and development can reduce costs and provide customer satisfaction while generating information that can be analyzed using big data techniques [59]. Data generated by IoT devices generates huge amounts of outsized information that must be stored properly and analyzed in real-time in order to make decisions. Deep learning provides high levels of accuracy when analyzing massive amounts of information [60]. To develop a high-tech society, IoT, Big Data Analytics, and Deep Learning will serve as a strong foundation.

CONCLUSIONS

Research and developers have recently become interested in IoT's recent advancements. Researchers and developers at the forefront of the Internet of Things are playing a key role in increasing technology on a large scale while simultaneously benefiting society to the fullest extent. However, improvements are possible as long as we consider the varied issues and shortcomings within the present technical approaches. A number of problems and challenges were described in this survey article, aimed at helping those who develop and research to build a more effective model and identifying important application areas for IoT developers and researchers. It is important to recognize that IoT is not only providing services but also generating a tremendous amount of knowledge. Hence, massive data analytics may be able to give the data necessary to develop an improved IoT system.

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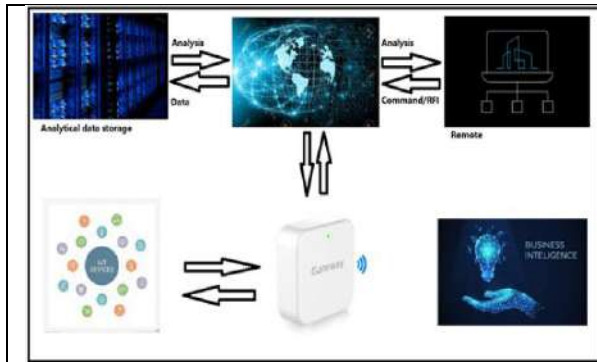
Fig.1. Architecture of IoT
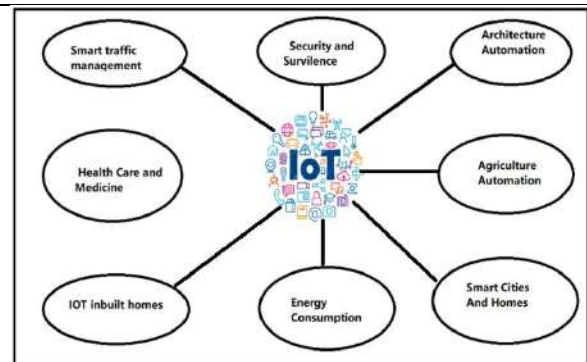
Fig.2 Applications of IoT
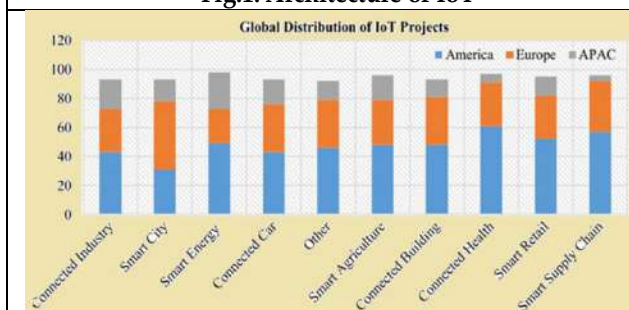
Fig.3 Global implementation of IoT projects in America , Europe and APAC
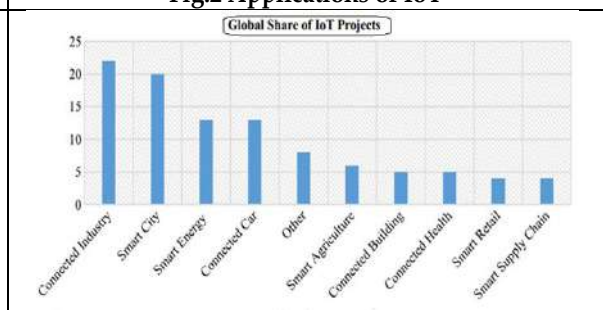
Fig.4 Global share of IoT projects
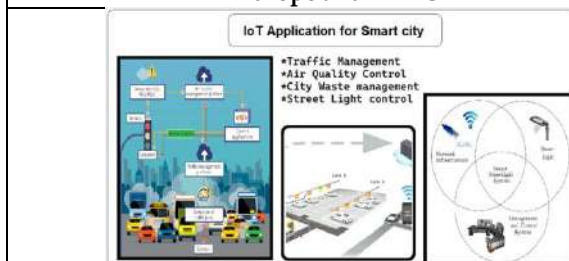
Fig.5 IoT Application for Smart City
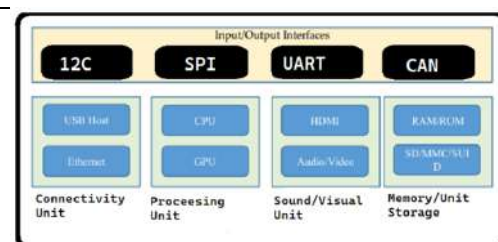
Fig.6. Function module of IoT system

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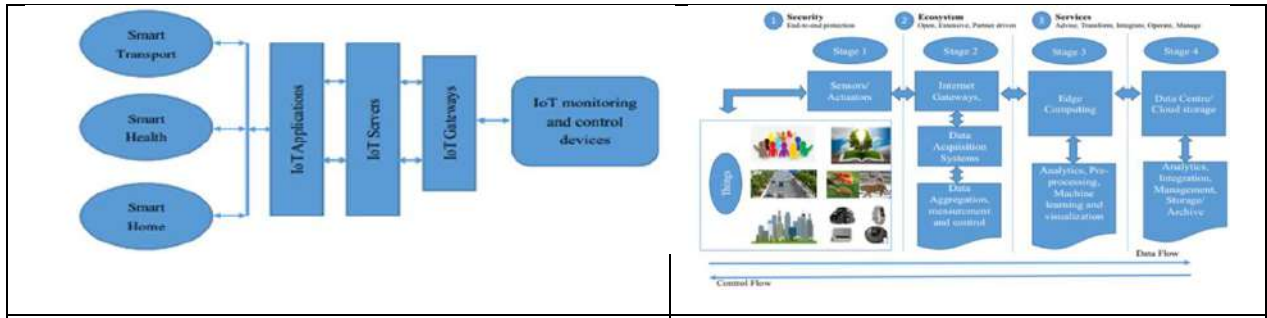
| Fig.7. Structure of IoT | Fig.8. Stages of IoT |

RESEARCH ARTICLE

A Drug Review on Efficacy of "Vallarai Nei" - A Siddha Herbal Formulation in the Management of Pitha Moothira Kiricharam (Urinary Tract Infection)

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ABSTRACT

Urinary tract infections (UTIs) are some of the bacterial infections, commonly affecting 150 million people every year worldwide and also the most common outpatient infections with a peak in young women aged 14–24 years old, the prevalence of UTIs increases with age. In health, bacterial colonization is confined to lower end of the urethra and the remainder of the urinary tract is sterile. According to *Yugi Vaithiya Chinthaamani-* a siddha classical text, Urinary disorders are classified into two major groups depending upon the symptoms. They are *Neerinai Arukkal Noi* and *Neerinai Perukkal Noi*. *Moothira kiricharam* was described as one of the types in *Neerinai Arukkal Noi*. *Moothira kiricharam* is further divided into 4 types, in which the causes, signs and symptoms of *Pitha Moothirakiricharam* could be correlated with Urinary tract infection in modern system. The urinary tract can become infected with range of pathogens, but most commonly by *Escherichia coli*, *Klebsiella pneumoniae*, *Proteus mirabilis*, *Enterococcus faecalis* and *Staphylococcus saprophyticus*. UTI typically occurs by an uropathogen residing in the gut which contaminates periurethral region followed by colonization of the urethra and subsequent migration of the pathogen to the bladder, with the help of appendages such as flagella and pili. Multiple bacterial adhesions recognize receptors on the uroepithelium (bladder epithelium) and mediate colonization. The keystone of treatment for any bacterial infection, including a UTI, is antimicrobial therapy. Antimicrobial resistance is rapidly increasing along with the development of classical antibiotics accordingly, there is an urgent need to develop a different drug line to treat and control Multi Drug Resistant bacterial infections. Plant derived antimicrobial substances are plant-originated secondary metabolites and have

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great concern because of their antibiotic activity without conferring resistance. This is why the people switch over to the traditional system of medicine. In Siddha System of medicine large number of drugs has been used for the management of UTI and to prevent its recurrence. This review article focuses on one such polyherbal Siddha formulation '*Vallarai Nei'* which is cost effective, efficacious and easily available that is mentioned in text, *Chikicharathna Deepam Vaithiya Sindhamani*, specifically for Urinary tract infection.

Keywords: Vallarai Nei, Moothira kiricharam, *E.coli*, Urinary tract infection, antimicrobial.

INTRODUCTION

Siddha system of medicine, have listed the diseases of mankind as 4448 based on the alteration in three humours (Mukkutram) -*Vali, Azhal, Iyyam* [1]. According to *Yugi Vaithiya Chinthaamani-* a siddha classical text, Urinary disorders are classified into two major groups depending upon the symptoms. They are *NeerinaiArukkal Noi* and *Neerinai Perukkal Noi* [2]. *Moothira kiricharam* was described as one of the type in *Neerinai Arukkal Noi. Moothira kiricharam* is further divided into 4 types, in which the causes, signs and symptoms of *Pitha Moothira kiricharam* could be correlated with Urinary tract infection in modern system [2,3]. Urinary tract infections (UTIs) are some of the bacterial infections, commonly affecting 150 million people every year worldwide [4]. They are also the most common outpatient infections with a peak in young women aged 14–24 years old, the prevalence of UTIs increases with age. Urinary tract infection is the term used to describe acute urethritis and cystitis caused by microorganism. In health, bacterial colonization is confined to lower end of the urethra and the remainder of the urinary tract is sterile [5] . The urinary tract can become infected with range of pathogens, but most commonly by *Escherichia coli*, *Klebsiella pneumoniae*, *Proteus mirabilis*, *Enterococcus faecalis* and *Staphylococcus saprophyticus* [6]. UTIs are grouped as uncomplicated or complicated. Uncomplicated UTIs typically affect individuals who are healthy and have no structural or neurological urinary tract abnormalities, in which these infections are differentiated into lower UTIs (cystitis) and upper UTIs (pyelonephritis) [4]. The prevalence of urinary tract infection is about 3% in women at the age of 20 which is getting increase by about 1% in each successive decade. It is estimated that 20% or more of the female population suffers from some form of UTI in their lifetime. Adherence is a key event initiating each step in UTI pathogenesis. A UTI typically occurs by an uropathogen residing in the gut which contaminates periurethral region followed by colonization of the urethra and subsequent migration of the pathogen to the bladder, with the help of appendages such as flagella and pili [4]. Multiple bacterial adhesins recognize receptors on the uroepithelium (bladder epithelium) and mediate colonization. Uropathogens such as UPEC survive by invading the bladder epithelium, producing toxins and proteases to release nutrients from the host cells. By multiplying and overcoming host immune surveillance, the uropathogens can subsequently ascend to the kidneys, again attaching via adhesins or pili to colonize the renal epithelium and then producing tissue-damaging toxins. Consequently, the uropathogens can cross the tubular epithelial barrier which results in bacteraemia [4]. Recurrent urinary tract infection (UTI) might be one of the most common problems in urological clinics. Recent research has exposed novel evidence about recurrent UTI and it should be considered as different disease from the first infection.

The pathogenesis of recurrent UTI might involve two mechanisms, bacterial factors and lack of host defense [6]. Bacterial survival in the urinary bladder after antibiotic treatment and progression to form intracellular bacterial communities might be the most important bacterial factors. In deficiency of host defense, a defect in pathogen recognition and urothelial barrier function impairment plays the most important roles. Immunodeficiency and urogenital tract anatomical abnormalities have been considered the essential risk factors for recurrent UTI. In healthy women, voiding dysfunction and behavioral factors also enhance the risk of recurrent UTI [7]. Recurrent infection may lead to significant morbidity. If get complicated it may cause end stage renal failure. So it is need of the hour for earlier detection and healing of this infectious disease among the population.

The keystone of treatment for any bacterial infection, including a UTI, is antimicrobial therapy. Since UTIs are very common in women, the initiation of antibiotics usage for treatment is a vital one. Antimicrobial resistance is rapidly increasing along with the development of classical antibiotics accordingly, there is an urgent need to develop a different drug line to treat and control MDR bacterial infections. Medicinal values of plants were known to earlier traditional medical practitioners. Plant derived antimicrobial substances are plant-originated secondary metabolites and have great concern because of their antibiotic activity without conferring resistance [8]. This is why the people switch over to the traditional system of medicine. In Siddha System of medicine large number of drugs has been used for the management of UTI and to prevent its recurrence. Siddha formulations not only treat the disease but also strengthen the urinary tract and correct the deranged *pitham* without any undesirable side effect. This review article focuses on one such polyherbal Siddha formulation '*Vallarai nei*' as mentioned in text *Chikicharathna Deepam Vaithiya Sindhamani*, specifically indicated for Urinary tract infection [9] which is cost effective, efficacious and easily available.

MATERIALS AND METHODS

Ingredients of Vallarai Nei (Medicated ghee of Vallarai) [9]
PART A:

- Vallarai Charu (*Centella asiatica*. Linn)   - 1.34 litre (1 padi)
- Poduthalai Charu (*Phyla nodiflora.*Linn)   - 1.34 litre (1 padi)
- Ponangani Charu (*Alternantherasessilis.*Linn)   - 1.34 litre (1 padi)
- Elumichai Charu (*Citrus limon.*Linn)   - 0.67 litre (1/2 padi)
- Aavin paal (*Cow's milk*)   - 2.68 litre (2 padi)
- Aavinnei (*Cow's ghee*)   - 2.68 litre (2 padi)

PART B

- Sathikkai (*Myristica fragrans.*Linn)
- Sathipathiri (*Myristica fragrans.*Linn)
- Masikkai (*Quercus infectoria.*Linn)
- Karkadakashingi (*Rhus succedanea.*Linn)   each
- Athimathuram (*Glycyrrhiza glabra.*Linn)   10.2 gram
- Val milagu (*Piper cubeba.*Linn)   (2 kalanju)
- Kattathi poo (*Woodfordia fruticosa.*Linn)
- Elam (*Elettaria cardamomum.*Marton)
- Kirambu (*Syzygium aromaticum.*Linn)

Method of Purification [9]

1. The leaves of Vallarai (*Centella asiatica.Linn*), Poduthalai (*Phyla nodiflora.Linn*), Ponnanganni (*Alternanthera sessilis.* Linn) should not be washed but cleaned with white cloth and remove the rotten and ripen leaves.
2. Sathikkai (*Myristica fragrans.Linn)* - the outer skin of the fruit was peeled off and made into small pieces and dried under the sunlight.
3. Sathipathiri *(Myristica fragrans. Linn),* Masikkai (*Quercus infectoria.Linn*) and Elam (*Elettaria cardamomum. Marton,* Kirambu*(Syzygium aromaticum.* Linn*)* are cleaned and dried under the sunlight.
4. Karkadakasingi*(Rhus succedanea.Linn*) - Fried in badam oil and taken.
5. The roots ofAthimathuram (*Glycyrrhiza glabra.Linn)* arewashed with clean water then the skin is pealed and made into small pieces.
6. Valmilagu*(Piper cubeba.Linn)* - The stalk is removed and dried in the sunlight

7.   Kattathi poo (*Woodfordia fruticosa.Linn*) - The leaf, stem are removed and dried under the sunlight

Method of preparation [9]

The leaf extracts along with lemon juice then cow's ghee and milk mentioned in the part A are taken in a pot and heated gently. Then the raw drugs mentioned in part B are made into a fine powder (chooranam) and grinded with milk and added to the above mixture. And when the required consistency is reached filter the content and add 70 gram of sugar. This prepared medicine was stored in air tight glass container and labelled.

Drug Storage

The study drug Vallarai Nei is stored in a cleaned and dried wide mouthed glass bottle.

Dose & Duration [9]

Dose: Uchikarandi – 16 ml,before food with milk/warm water for 15 days.

VALLARAI

Botanical Name: *Centella asiatica*.Linn
English Name: Indian Pennywort
Family: Apiaceae (Umbelliferae)
Part used: Whole plant
Taste: Sour, Bitter, Sweet
Character: Coolant
Division: Pungent
Action: Alterative, Tonic, Diuretic, Stimulant, Emmenogogue [10]

Phytochemical constitutents:

Aglycoside, Asiaticoside, Brahmoside, Brahminoside, Centic acid, Centellic acid, Indocentoic acid and Stigmasterol [11].

Pharmacological activity

Antistress, Antiepileptic, Antiulcer, Anti microbial, Anti viral, Anti cancer, Cardio protective, Wound healing and Spasmolytic [12,13]

Medicinal uses

The plant is valued in indigenous medicine for treatment of leprosy and skin disease and to improve memory.
▪   The cold poultice of the fresh herb is used as an external application in rheumatism, elephantiasis, hydrocele.
▪   Internally it is given as tonic and is used in bronchitis, asthma, gastric catarrh, leucorrhea, kidney troubles, urethritis and dropsy.
▪   The leaf extract is used in the preparation of medicated oil for bone fracture. The leaves are diuretic [13].

Scientific review

In agar diffusion assay, the methanol leaf extract of *Centella asiatica* showed the broad-spectrum antibacterial activity against both gram positive and gram-negative organisms with zone of inhibition ranging 10mm to 19mm.The methanol leaf extract of *C.asiatica* has moderate antibacterial activity against *Staphylococcus aureus*, *Bacillus cereus, Pseudomonas aeruginosa* and *Escherichia coli,* while the acetone extract has less effective against these organisms[14].

PODUTHALAI

Botanical Name: *Phyla nodiflora.Linn*
English Name: Purple lippie**,** Frog fruit
Family: Verbenaceae
Part used: Leaves, fruit

Taste: Bitter, Astringent
Character: Hot
Division: Pungent
Action: Demulcent,Deobstruent, Diuretic, Astringent, Expectorant, Tonic[10]
Phytochemical constitutents:

Triterpenoids, Lippiacin, Nodifloridin, β-Sitosterol glycoside, Stigmasterol glycoside, Flavanoid, Phenols, Steroids [15,16]

Pharmacological activity:

Anti microbial activity, Anti fungal activity, Anti inflammatory activity, Anti tumour, Anti urolithiatic activity, Anti diabetic, Larvicidal activity[15,16,17]

Medicinal uses

- Traditional medicine reports that a poultice comprised of fresh plant is a cure for cervical glands.
- A mixture of Lippa nodiflora and seeds of *Cyminum cuminum* is used to treat gonorrhea
- The plant decoction along with Leucas aspera and roots of *Ocimumgratis simum* is known for its anti-malarial property [10].

Scientific review

The methanol extract of *Phyla nodiflora* had been assessed for antibacterial (*S.aureus, M.luteus, P.mirabilis*) which was performed by agar diffusion method using a paper disc. The anti-microbial activity was seen in the essential oils of this plant against *E.coli, P.aeruginosa* and *Stapylococcus aureus*. The antibacterial activity was shown by the seeds (methanolic extract) of this plant also [15,17].

PONNANGKANI
Botanical Name: *Alternanthera sessilis.* **Linn**
English Name: Sessile plant
Family: Amaranthaceae
Part used: Whole plant
Taste: Sweet
Character: Coolant
Division: Sweet
Action: Alterative, Coolant[10]
Phytochemical constituents:

2,4-methylenecycloartanol, Cycloeucalenol, Choline, Oleanolic acid, Saponins, Lupeol 117, β-Sitosterol, Campesterol, α-tocopherol [18,19]

Pharmacological activity

Anti microbial activity, Anti oxidant activity. Anti pyretic activity. Nootrophic activity, Wound healing activity, Hepato protective activity, Anti ulcer activity, Hematinic activity, Anti diabetic activity[19,20]

Medicinal uses

- The sessile leaves is fried with ghee and eaten to produce cooling effect to eyes.
- The leaves are very potent in treating cuts and wounds, used as an antidote for scorpion sting and snake bite
- The whole plant when dried it is used for purifying blood and cures skindiseases [19].

Scientific Review

The ethanolic extract of *A.sessilis* showed maximum zone of inhibition against *Staphylococcus aureus* and *Staphylococcus hemolyticus* and exhibit antifungal activity against *Candida albicans*[18].

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ELUMICHAI
Botanical Name: *Citrus limon.*Linn
English Name: Lime
Family: Rutaceae
Parts used: Leaves, Fruit, Fruit juice, Essential oil
Taste: Sour
Character: Hot
Division: Pungent
Actions: Refrigerant [10]

Phytochemical constituents
Eriodictyol, Hesperidin, Hesperetin , Apigenin, Diosmin, Quercetin, Limocitrin, Spinacetin, Orientin, Vitexin, Ferulic acid, Synapic acid, geranial, myrcen, Limonoids, p-hydrozybenzoic acid [13,21,22].

Pharmacological activity:
Anti cancer, Anti oxidant, Anti-inflammatory, Anti microbial, Anti parasitic, Anti allergic, Hepato-regenerating, Anti diabetic, Anti obesity[13, 21,22]

Medicinal uses
- It is very rich in Vitamin C, so it has potassium and calcium which possesses anti scorbutic activity
- Lemon has high citric acid content which has the property to dissolve or to stop the development of small kidney stones.
- It is also used as a preventive medicine for cholecystitis.
- Lemon consumption or even inhalation of the aroma has good effect on mood and tension, nervousness, anxiety, exhaustion and fatigue[21].

Scientific Review
- Acetone extracts from *C.limon* fruits have shown inhibitory activity against the Gram positive bacteria *Enterococcus faecalis* and *Bacillus subtilis*.
- Another study confirmed that *C.limon* essential oil promoted a 100% reduction in the growth of *C.albicans*[21].

SATHIKKAI
Botanical Name:*Myristica fragrans*Houtt**.**
English Name: Nut meg
Family: Myristicaceae
Parts used: Fruit
Taste: Pungent, Astringent
Character: Hot
Division: Pungent
Actions: Stimulant, Carminative, Narcotic, Aromatic, Aphrodisiac, Tonic[10]

Phytochemical constituents:
Myristicin, Elemicin, Camphene, Eugenol, Isoelemicin, Methoxyeugenol, Volatile oil, Sabinene, α-pinene.24,25

Pharmacological activity
Anti oxidant, Analgesic, Anti septic, Anti carcinogenic, Anti convulsant, Anti inflammatory, Anti bacterial, Anti fungal, Anti diabetic[24,25]

Medicinal uses
- It is used as folktale medicine to treat diarrhoea, sleeplessness, mouth ulcers

- The essential oil of nutmeg is used externally for rheumatism
- It prevents hypercholesterolemia and atherosclerosis. It is also useful as a tonic for brain and heart.[24]

Scientific review
- The extract of *M.fragrans* showed antibacterial activity against *Staphylococcus aureus*, *Proteus vulgaris* and *Klebsiella pneumonia*[25]

CHATHI PATHTHIRI
Botanical Name: *Myristica fragrans*Houtt**.**
English Name: Arillus of the Nut
Family: Myristicaceae
Parts used: Aril
Taste: Pungent, Astringent
Character: Hot
Division: Pungent
Actions:
Aphrodisiac, Carminative, Stimulant, Hypnotic[10]

Phytochemical constituents
Sabinene, α-pinene, Mycrene, Limonene, 1,8-cineole, Terpenol-4-ol, Myristicin, γ-terpinene, Malabaricone B[25]

Pharmacological activity
Anti inflammatory, Analgesic, Anti convulsant, Anti depressive, Anti cholinergic, Anti bacterial, Anti fungal, Anti diabetic[25,26]

Medicinal uses
- Mace yields essential oil used for flavoring food products and it is recommended for inflammation of bladder and urinary passage[26]
- The major benefits of the essential oil were it eradicates the bad breath, provides relief from sleeping disturbances and it is good for indigestion.[25]

Scientific Review
- The methanol extract of *M. fragrans* arils (mace) have good effect against *Candida albicans* (0.237 mg/ml). The two antimicrobial Rescorinols Malabaricone B and malabaricone C from *M.fragrans* mace exhibits strong antifungal and antibacterial.[26]

ELAM
Botanical Name:*Elettaria cardamomum* (L.) Maton
English Name: Cardamom seeds
Family: Zingiberaceae
Parts used: Seeds
Taste: Pungent
Character: Hot
Division: Pungent
Actions:
Stimulant, Carminative, Stomachic[10]
Phytochemical constitutents:
Geraneol, Terpineol, Mycrene, Hepatone, Camphor, Cineol, Trans nerodiol, Farsneol, Limonene, Sabinene[27,28]

Pharmacological activity
Anti fungal, Anti bacterial, Cardio protective, Anti diabetic, Hypocholesterolemic activity, Anti diarrheal activity, Anti caner activity and Anti ulcerogenic activity[27,28]

Medicinal uses
- It is used to cure sinusitis and head disorders.
- Cardamom powder is used for cough, dryness of mouth, abdominal pain
- In traditional medicine, consuming cardamom along tablespoon of honey daily improves the eyesight.
- Cardamom seeds are chewed to avoid bad breath, vomiting and indigestion.[28]

Scientific review:
The methanolic and ethanolic extracts of cardamom showed inhibition against the isolates of *S.aureus* and *P.mirabilis*. The essential oils of cardamom shows inhibition against *C.albicans* and *S.mutans* with maximum zone of inhibition 11.6 0.56mm and 11.4 0.55 mm followed by S.aureus with 9.8 0.20 mm zone of inhibition and also effective against E.coli and Streptococci mutans [27].

MACHIKKAI
Botanical Name: *Quercus infectoria*. Olivier
English Name: Magic nuts, Oak galls
Family: Fagaceae
Parts used: Fruit
Taste: Astringent
Character: Coolant
Division: Pungent

Actions:
Styptic, Astringent, Tonic[10]
Phytochemical constitutents:
50-70% of Gallo tannic acid,Gallic acid, Ellagic acid, Sitosterol, Methyl betulate, Methyl oleanolate, Nyctanthic acid, Roburic acid [29].

Pharmacological activity:
Anasthetic, Analgesic, Anti inflammatory, Anti microbial, Anti oxidant, Anti cancer Anti diabetic and Anti hypertensive[29]

Medicinal uses
- It is used as one of the most powerful astringent in siddha medicine. Its powder is utilized in a number of tooth powders for strengthening teeth and gums.
- The oak gall decoction is used to gargle for sore throat, stomatitis, tonsillitis.
- Powdered galls are given internally for the treatment of leucorrhea and other vaginal discharges.

Scientific review
In vitro antibacterial activity of methanol and aqueous extract of *Quercus infectoria* galls against several bacterial pathogens of the urinary tract infection was evaluated using disc diffusion method at the concenteration of 5 mg/disc. Both the extracts showed similar inhibitory effect against *Staphylococcus saphrophyticus*, *Streptococcus agalactiae*, *Streptococcus pneumonia* and *Enterococcus faecalis* (Gram positive bacteria) and against *Proteus mirabilis* (Gram negative bacteria). It has also been reported to be effective against *E.coli, Staphylococcus aureus, Psuedomonas aeruginosa and Bacilus subtilis*. The ethanol extract showed maximum inhibition against *E.coli, Staphylococcus aureus, Psuedomonas aeruginosa* and *Candida albicans*.[29]

ATIMADURAM

Botanical Name:*Glycyrrhiza glabra* L.
English Name: Jequility: Indian or Jamaica liquorice
Family: Fabaceae
Parts used: Roots
Taste: Sweet
Character: Coolant
Division: Sweet

Actions:

Emolient, Demulcent, Mild expectorant, Laxative, Tonic[10]

Phytochemical constituents:

Glycyrrhizin, Licoagnone, Glucoside, Liquoric acid, Glycyrretol, Glabrone, Glyzarin, Glabridin, Liqcoumarin, Liquirtin[30,31]

Pharmacological activity:

Anti inflammatory, Anti oxidant, Expectorant, Anti bacterial, Anti allergic, Anti fungal, Anti carcinogenic, Hepato protective and Anti malarial [30,31].

Medicinal uses:

- The dried roots have been used as expectorant and carminative by the Indian peoples.
- Leaves of liquorice were used externally to treat cuts and wounds
- It is useful in ulceration of urinary tract, polydipsia and cough.[10,30]

Scientific review

The alcoholic extract obtained by percolation from roots of *Glycyrrhiza glabra* showed maximum inhibition against *E.coli, E.faecalis* with inhibition diameter of 15 mm. The methanolic extract exhibited potent antimicrobial activity against *Staphylococcus aureus* with a zone of inhibition of 22 mm. The antifungal activity of hydroalcoholic extract prepared from rhizomes and roots of *G.glabra* showed inhibitory zones at the tested concentration (50mg/ml) after 24 hour, were 1.0 – 1,2 cm for *Candida albicans* using the disc diffusion halo assay.[31]

KIRAMBU

Botanical Name: *Syzygium aromaticum* (L.) Merr.&L.M.Perry
English Name: Cloves
Family: Myrtaceae
Parts used: Flower buds
Taste: Pungent
Character: Hot
Division: Pungent

Actions:

Antispasmodic, Carminative, Stomachic[10]

Phytochemical constituents:

Eugenol, Isobiflorin, Acety leugenol, Caryophyllene, Eugenine, Eugenone, Syzyginis, Eugenol acetate, Napthalene[32]

Pharmacological activities:

Anti fungal, Anti bacterial, Anti oxidant, Anti inflammatory, Anti carcinogenic, Anesthetic, Analgesic and Anti thrombotic[32]

Medicinal uses
- The cloves are stimulant and carminative which is used for treating dyspepsia and gastric irritation
- Essential oil called clove oil is used as an ingredient of dentifrices, gargles and chewing gums.
- It is used as a local analgesic for hypersensitive dentines and caries cavities.[32]

Scientific Review
In vitro evaluation of antibacterial activity of clove was screened against several uropathogens. The best antibacterial activity of Clove was demonstrated against *E.coli* and *Proteus* species as they had maximum zone of inhibition and it has significant antibacterial activity against *K.pneumoniae*. Clove oil was found to be effective against gram negative bacteria like *E.coli, P.aeruginosa*.[32]

VALMILAGU
Botanical Name: Piper cubeba L.f.
English Name: Cloves
Family: Myrtaceae
Parts used: Unriped fruit
Taste: Pungent
Character: Hot
Division: Pungent

Actions
Stimulant, Carminative, Diuretic, Expectorant[10]

Phytochemical constituents
Sabinene, α-thujene, Carene, Caryophyllene, Copaene, α and β cubebene, Germacrene1,4, 1,8 – Cineole, Cubebol[33]

Pharmacological activity
Anti microbial, Anti inflammatory, Analgesic, Anti mutagenic, Anti oxidant[33]

Medicinal uses
- Fruits called cubebs, used as a spice and in medicine for dysentery and as a local irritant and diuretic
- They are used to treat rheumatism, gonorrhea and bronchial troubles
- It also has an inhibitory effect on hepatitis C virus protease.
- The dried cubebs are taken internally for oral and dental diseases, halitosis and for hoarseness of voice.[33]

Scientific review
The acetonic and ethanolic extracts of *P.cubeba* showed a greater antibacterial activity against *S.aureus* (18.96 mm and 17.32mm respectively) whereas the aqueous extract showed greater antibacterial activity on gram negative *E.coli* (15mm) and *P.aeruginosea* (15 mm). This is due to the presence of phenol and flavonoids in the aqueous extracts.[33]

KATTATTI
Botanical Name: *Woodfordia fruticosa* L.f.
English Name: Fire-flame bush
   Shitanjitea Wood fordia,
   Bauhini tomentos
Family: Lytheraceae
Parts used: Leaves, buds, flowers, tender fruit, Riped fruit
Taste: Astringent
Character: Coolant
Division: Sweet

Actions:
Antidysenteric, Antiheminthic [10]

Phytochemical constituents:
β Sitosterol, Octacosanol, Hecogenin, Meso-inositol, Ellagic acid, 3-rhamnoside, 3- β-1-arabinoside, Pelargonidin 3,5-diglucoside, Cyanin 3,5-diglucoside, Telllimagrandin, Oenothein A, Oenothein B, Woodfordin A-D [34,35]

Pharmacological activity
Anti microbial, Hepatoprotective, Cardioprotective, Antioxidant, Antiulcer, Antifertility, Anti hyperglycemic, Anti hyperlipidemic and Anti inflammatory [34]

Medicinal uses
- The flower of this plant is used to treat leprosy, leucorrhea, fever, toothache, dysentery.
- The dried flowers are used to treat sprue, bowel disease and infertility
- It is powdered and dusted over ulcers and wounds to reduce discharge.
- It is used as a substituent for Glycyrrhiza glabra according to Yogaratnakara.[34]

Scientific review
The methanolic extract of fruticosa flower was more effective against gram negative bacteria as compared to gram positive bacteria. Against 14 human pathogens, different extracts of dried flowers of *W.fruticosa* evaluated for their significant antimicrobial activity. Out of these extracts tested, petroleum ether extract showed significant activity when compared to Gentamicin as standard drug.[34]

KARKADAGA SHINGI
Botanical Name: *Rhus succedanea* L.
English Name: The galls
Family: Anacardiaceae
Parts used: Galls
Taste: Astringent
Character: Hot
Division: Pungent

Actions:
Astringent, Tonic, Nutritive, Digestive, Expectorant, Stimulant, Cholagogue.[10]

Phytochemical constituents:
Tannins, Phenolic acids, Anthocyanins, Gallic acid, Flavonoid glycosides and Organic acids[36]

Pharmacological activity
Anti inflammatory, Anti oxidant, Anti malarial, Anti mutagenic, Anti microbial, Antifungal, Anti viral and Hypoglycemic [36].

Scientific Review
The methanol extract of galls of R.succedanea was most effective against gram-negative bacteria (*E.coli*) whereas the aqueous extract of crude drug showed higher inhibition against Gram- negative bacteria such as *E.coli* and *P.aeruginosa* [36]

AAVIN NEI – COW'S GHEE:
Synonyms: Ko nei, Pasunei, Thuppu[37]
Pharmacological activity:

- Anti oxidant
- Anti inflammatory
- Anti viral
- Tonic
- Anticancer
- Hepato protective
- Wound healing property[39]

Phytochemical constituents:
- Vitamin A 3500/100gm
- Triglycerides
- Di-glycerides
- Mono-glycerides
- Phospholipids
- Beta-carotene 600 IU
- Vitamin E.[39]

Medicinal uses
- Cow ghee also known as the clarified butter, obtained from the cow milk has been known for many medicinal properties like it is rejuvenating, bestows luster and beauty, enhances memory and stamina, increases the intellect and promotes longevity.
- It has about 8% of saturated fatty acids which make it easily digestible.
- It is an aphrodisiac and protects the body from various diseases.
- It has lubricating property which is helpful in reducing the symptoms of CVS.
- Cow ghee is excellent in balancing Vatham and Pitham related diseases.
- It is better than any other animal or vegetable fat since it has 96% rate of absorption.[39]
- Cow's ghee is used for excessive thirst, vomiting, ulcer, sexually transmitted diseases,hiccups, cough, burning sensation of stomach, bone diseases, piles and as antidote.[37]

Aavin Paal – Cow's milk:

Synonyms: Bayam, Keeram, Suthai, Payasu, Paagu, Amuthu, Thutham, Chaaru[37]

Bionutrients in cow's milk:
- Cow's milk is composed of about 87% water, 3%–4% fat, 3.5% protein, about 5% lactose, and 1.2% minerals.
- Most of the fatty acids in milk are saturated fatty acids (60%) which are mainly represented by palmitic acid, followed by myristic and stearic acids
- Carbohydrates in milk are almost represented by lactose, a disaccharide, which is cleaved into glucose and galactose by the intestinal enzyme lactase and made it available for absorption
- Caesins contribute about 80% of the protein fraction in cow's milk which mostly contain glutamic acid, proline, arginine and branched amino acids such as leucine, isoleucine and valine.
- Milk provides many minerals, in particular calcium and phosphorus, but also potassium, zinc magnesium, , and selenium. It also provides both B-group water-soluble vitamins (riboflavin and B12) and fat-soluble vitamins (e.g., A and E)
- Milk proteins are of high nutritional value since they contain all the essential amino acids required by the human body which are highly digestible and bioavailability one.[40,41]

Actions:
- Anti diabetic
- Anti hypertensive
- Anticholesteremic
- Anti microbial

- Cardio protective
- Chemoprevention
- Improvement of cognitive function[41]

Antimicrobial activity of milk:

A number of proteins found in milk under various conditions exhibit antimicrobial activity which includes lactoferrin, lactoperoxidase, lysozyme and possibly N-acetyl-ß-D-glucosaminidase. Lactoferrin is one of the minor proteins naturally occurring in cow milk at an average concentration of about 0.2 grams/liter. It has been shown that 'natural' lactoferrin is bacteriostatic against a wide range of micro organisms, including gram-positive organisms such as *Staphylcoccus aureus* and bacillus species, Lactoperoxidase itself has no antibacterial activity. The antibacterial property of the lactoperoxidase system is based upon inhibition of vital bacterial metabolic enzymes brought on by their oxidation by hypothiocyanate.

Lysozyme possesses antibacterial activity against a number of bacteria. Cow milk may contain both c- and g lysozymes because both types are found in various other body fluids and in the stomach tissue of the cow. This enzyme usually functions in association with lactoferrin or immunoglobulin A. Lysozyme is effective against *Escherichia coli* in concert with immunoglobulin A. N-acetyl-ß-D-glucosaminidase activity along with elevated lactoferrin may contribute to increased antibacterial property. NAGase inhibits *Pseudomonas aeroginosa* and *Staphylococcus aureus* [42].

DISCUSSION AND CONCLUSION

Siddha system of medicine has copious formulations which contain several compounds naturally that are found in herbs, shrubs, trees and spices that have been shown to possess antimicrobial activity and serve as a source of antimicrobial agents against several uropathogens. Urinary tract infections (UTIs) are one among the most frequent types of bacterial infections occurring both in the community and hospital background. The inappropriate use and abuse of antimicrobials increased the resistance in pathogens infecting urinary tract as well as in normal human bacterial flora. Exploring the medicine from traditional basis alternatively with multiple action and also reduced resistance is of highly vital in treating UTIs. Based on the reviews from various siddha texts, the herbal ingredients of *Vallarai Nei* are common drugs that are used in treating *Neerinai arugal noi.* In view of the above-mentioned pharmacological activities, most ingredients are found to possess anti-bacterial and anti-fungal activities against uropathogens such as *E.coli,K.pneumoniae, Staphylococcus* sp, *Pseudomonas aeruginosa* and *Candida albicans*. Further the presence of the wide range ofanti-oxidants and anti-inflammatory activates made it the drug of choice in the treatment of Pitha Moothira Kiricharam (Urinary tract infection). The ghee acts as a carrier of nutrients to be absorbed across the cell membrane. Thus the effectiveness and potency of a drug is further enhanced. With this established efficacy, the drug is easily available to prepare, cost effective and safer treatment – *Vallarai Nei* serves as a capable anti-microbial preparation for future research in the treatment of Urinary tract infection. Further clinical studies and statistical data analysis help in exploring this herbal Siddha formulation.

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RESEARCH ARTICLE

Covid-19 Impact Analysis for Social Media using LDA Algorithm

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ABSTRACT

Social media is one of the most popular media where data related to anything and everything is populated by everyone in the world. We will analyze the impact of COVID – 19 industries using social media. Tracking of internet, social media messages, tweets, posts etc. We can analyze in industries vise. In every industry post positive, negative, and neutral tweets. We find the tweets in authorized profile. Our ultimate aim is to develop a system to analysis the impact in industries from all social media tweets and posts. Then analyze the data form twitter and find the result.

Keywords: COVID-19, Latent Dirichlet Allocation, Data Mining, Sentiment analysis, Twitter data, Topic modelling

INTRODUCTION

Big data is basically used to describe the exponential growth as well as the availability of structured and unstructured data. The big data applies inductive statistics and concepts from nonlinear system identification to conclude laws of regressions, nonlinear relationships and casual effects coming from large data sets to show relationships, dependencies as well as to perform predictions of outcomes and behaviors. Social media is a key model of the velocity and variety which are associated with big data. Only Big Data Applications can enable organizations to manage these social conversations in real-time. Big data is usually associated to having volume, velocity, variety, variability and complexity which are also features of Social Media Analytics There are over a billion active users of social media network worldwide, many of whom are frequently active and can be connected by means of their smart phones and tablets. Social media indeed has become a main communication network in the

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daily lives of people around the world. Data in Social media comes from numerous sources .It is a great challenge to undergo the different processes like linking, matching, connecting, correlating relationships, hierarchies and multiple data linkages. This is how complex data can be and if not managed properly, they can spiral out of control. It is absolutely to discover more about exploring Big Data with social media analytics.

Proposed Work

In this project we analyze the impact of COVID – 19 in industries using the social media platform twitter. In this project we can use rapid miner tool. We connect the twitter to rapid miner and generate the post we store that can be in mongo DB.As one of trending social media platform is twitter, so we can use that in our project. Twitter is becoming a promising information source to get the most timely knowledge and news around us. In this project we can analysis the impact in industries. How impact the Indian industries. In before that the project is only analysis the one industry. So that only analysis impact in one industry through the twitter. This project is mainly focused on the impact of multiple industries in India. We can analysis the impact through the twitter. In this project we analysis the impact in the industries are,

• IT Industry
• Car Industry
• Health Industry
• Agriculture Industry

We collect tweets form twitter. And then we analysis that the tweet is positive, negative and neutral. So that we can easily find which industry is affect more. And finally we combine all the industries we analysis then we can generate the impact.

Covid Impact Analysis For Social Media Using Lda Algorithm
Latent Dirichlet Allocation

The LDA model is extended by sampling those weights from a Dirichlet distribution, the conjugate prior to the multinomial distribution. Depending on the frequency of terms in each cluster the topics are classified using LDA. The topic terms are obtained using cluster analysis, D-pattern and Inner pattern evolving algorithms as shown as follow

LDA assumes the following generative process for each document in a corpus:

1. Choose B ~ Poisson (ξ).
2. Choose θ ~ Dir (α).
3. for each of the B words b_n:
(a) Choose a topic Y_k Multinomial(θ).
(b) Choose a word b_n from $p(w_n|Y_k,\beta)$, a multinomial probability conditioned on the topic Y_k.

First, the dimensionality k of the Dirichlet distribution (and thus the dimensionality of the topic variable y) is assumed known and fixed. Second, the word probabilities are parameterized by a k × V matrix β where $\beta_{ij} = p\,(w^j = 1|y^i = 1)$, which for now we treat as a fixed quantity that is to be estimated. Finally, the Poisson assumption is not critical to anything that follows and more realistic document length distributions can be used as needed. Scheme 1 discusses the procedure of converting text documents into topics with LDA.

COVID-19 impact analysis using social media has a number of benefits especially for analyzing the trustworthiness of public generated data and to avoid spreading of misinformation about a particular issue. It also help to prevent the violent activities before it happens. Analyze the public opinion of all tweets related to a COVID19 impact issue. Tweets are collected from twitter by giving a specific parameter. Then perform sentiment analysis to categorize the data into positive, negative, neutral.

Make Connection with Twitter

The Twitter Connector allows you to easily access Twitter data direct from Rapid Miner Studio. The connector can search for phrases, tweets, or user profile-information.

1. Create a new process in Rapid Miner Studio, drag the Search Twitter operator into the Process view, and click on the operator. Click the Twitter icon in the Parameters view to open the Manage Connections dialog. You can also open the Manage Connections window through Tools > Manage Connections**.**

2. Click on Add connection in the lower left of the window, give a name to the new connection, and set Connection type to Twitter Connection:

Establishing twitter connection

3. Click on Create and select your new Twitter connection.

4. To the right of the access token field, click the button to request an access   token.

5. Click on Request access token to open the Twitter website in your browser. If you are not already logged into your Twitter account, you will have to do so now. You can manually copy the URL by clicking on Show URL instead.

Authenticate Rapid Miner via OAuth

6. Allow Rapid Miner to access your Twitter account by clicking on Authorize app.

7. Copy the access token shown on the next page.

8. Return to Rapid Miner Studio, enter the access token, and click Complete

9. While not required, we recommend testing your new Twitter connection by clicking the Test button at the bottom of the Manage Connections window.

10. Close the Manage Connections dialog by clicking on save all changes.

Make Connection With Mongo DB

Before you can use the Mongo DB connector, you have to configure a new Mongo DB connection. For this purpose, you will need the connection details of your database (host name, port, and database name).If your Mongo DB installation requires authentication, you will also need valid credentials.

1. Open the Manage Connections dialog in Rapid Miner Studio by going to Tools –Manage Connections**.**

2. Click on Add Connection in the lower left.

3. Enter a name for the new connection and select Mongo DB Connection as   the Connection Type.

4. Fill in the connection details of your Mongo DB instance:

5. The preconfigured port is the default port used by Mongo DB. Note that Mongo DB does not require user authentication by default. Optionally, you can test the new configuration by clicking on the Test button.

6. Click Save all changes to save your connection and close the Manage Connections window.

Extract Data From Twitter

With the Search Twitter operator you can find all tweets containing a specified phrase, along with those tweets` metadata.

1. Click on the Search Twitter operator in the Process view.

2. Select your Twitter connection from the connection drop down menu in the operator parameters and fill in the query field. The query is the phrase you will search Twitter for.

Modules Are:
▪ Information Technology Industry
▪ Agriculture Industry
▪ Car Industry
▪ Health Industry

Electronics And Information Technology: We modify parameters like this: Collect data from twitter the impact in IT industry.

Agriculture Industry: We modify parameters like this: We collect data from twitter the impact of agriculture industry.

Car Industry: We modify parameters like this:

Health Industry: We modify parameters like this:

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We collect data from twitter the impact Health Industries, The Screenshots for Extract data from twitter, filtering, Sentiment analysis and API connection for Health Analysis Industry

Sentiment Analysis
Analyze tweets for sentiment i.e whether they are Positive, Negative or Neutral.

Store Results Into Mongo DB
Drag the Data to JSON operator, and the Write Mongo DB operator into the Process view and connect the operators as shown in the following screen shot. Select your Mongo DB connection and enter a name for the new collection**.**

Retrive Data From Mongo DB
To retrieve data from mongo DB give the appropriate collection name in which the data is stored.

CALCULATE RESULT

The analysis results are viewed in numbers to understand how much count of data generated in each The final results are drawn like pie chart:

CONCLUSION

This system helps to identify the COVID – 19 impact in industries of user-generated data through social media. This also helps us know that how industries can be affect. In this system we need about any other industries, then that can be also we collect the data from twitter. In this system can be analysis the data. In future we will post this result in twitter. So that people can be known that the impact of COVID -19.

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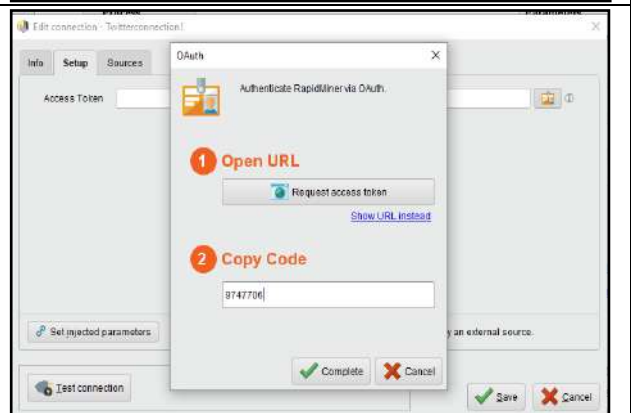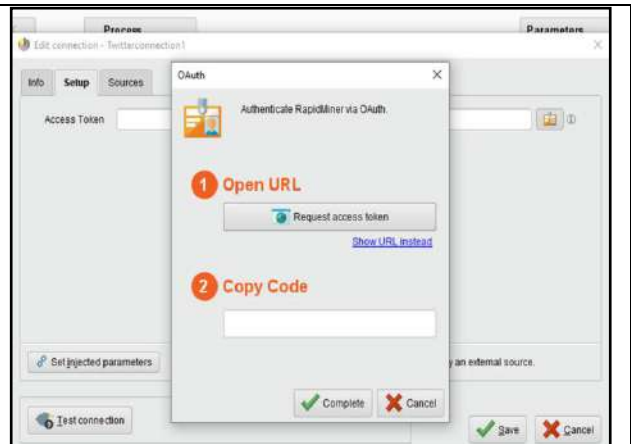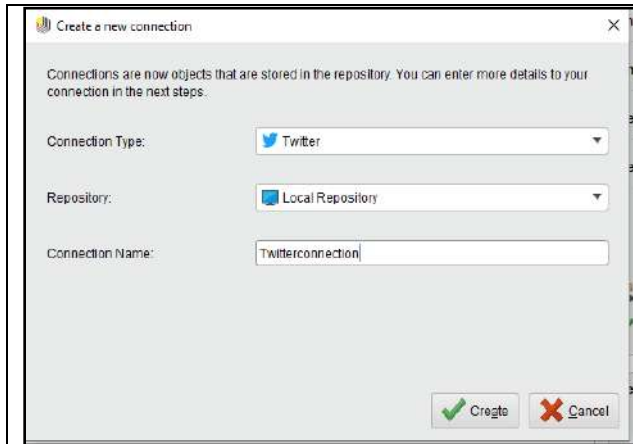
| Figure 4.4 Get access pin | Figure 4.5 Enter Access Pin |
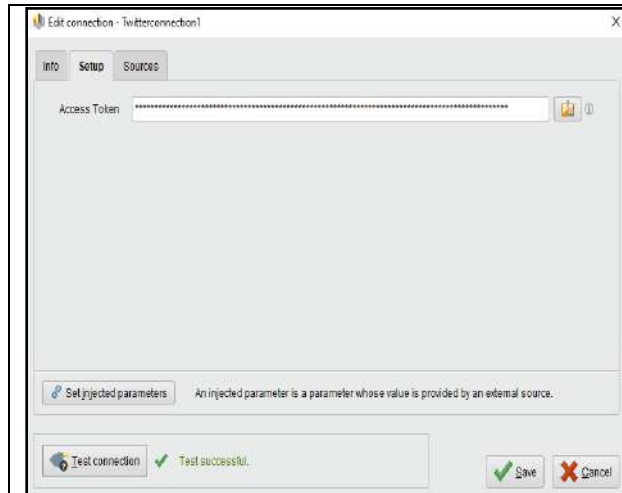
Figure 4.6 Enter access token	Figure 4.7 Establish new mongo DB connection
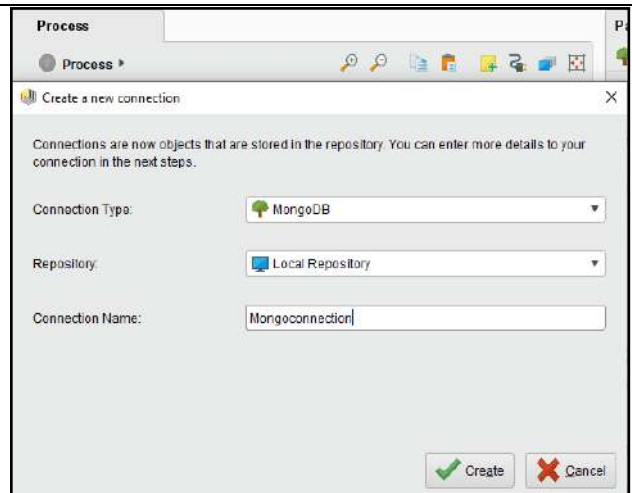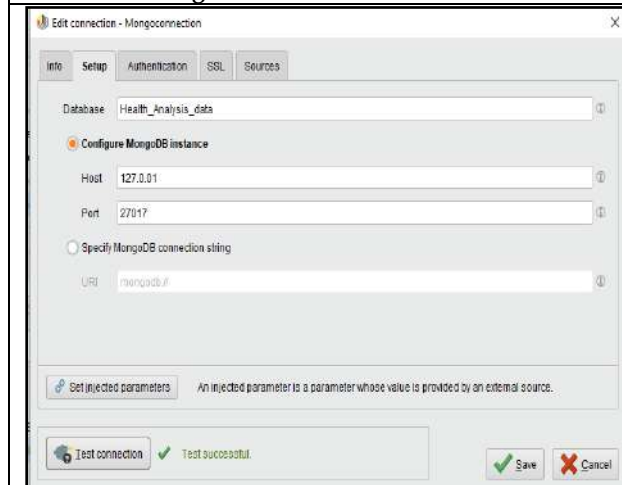
Figure 4.8 Configure mongo DB

Figure 4.9 IT Industry data

Figure 4.10 Agriculture Department data

Figure 4.11 Car Industry data

Figure 4.12 Health Analysis Industry

Figure 4.13 Filtering examples

Figure 4.14 Sentiment Analysis

Figure 4.15 Aylien Text Connection

Figure 4.16 Data to JSON

Figure 4.17 Write mongo DB

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Figure 4.18 Sentiment Analysis
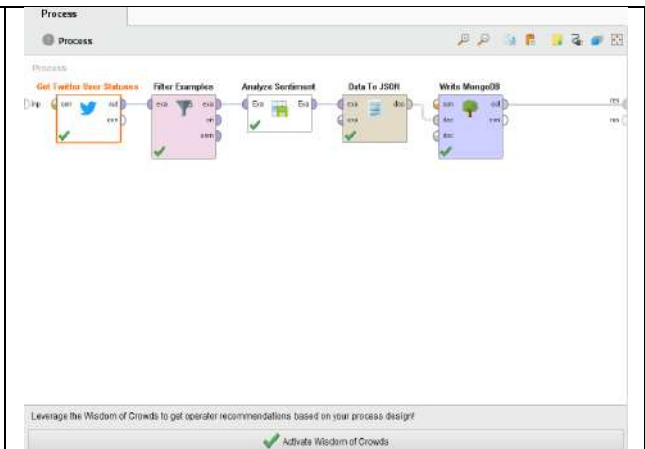
Figure 4.19 Write results in mongo DB

Figure 4.20 Read mongo DB

Figure 4.21 Result in table format
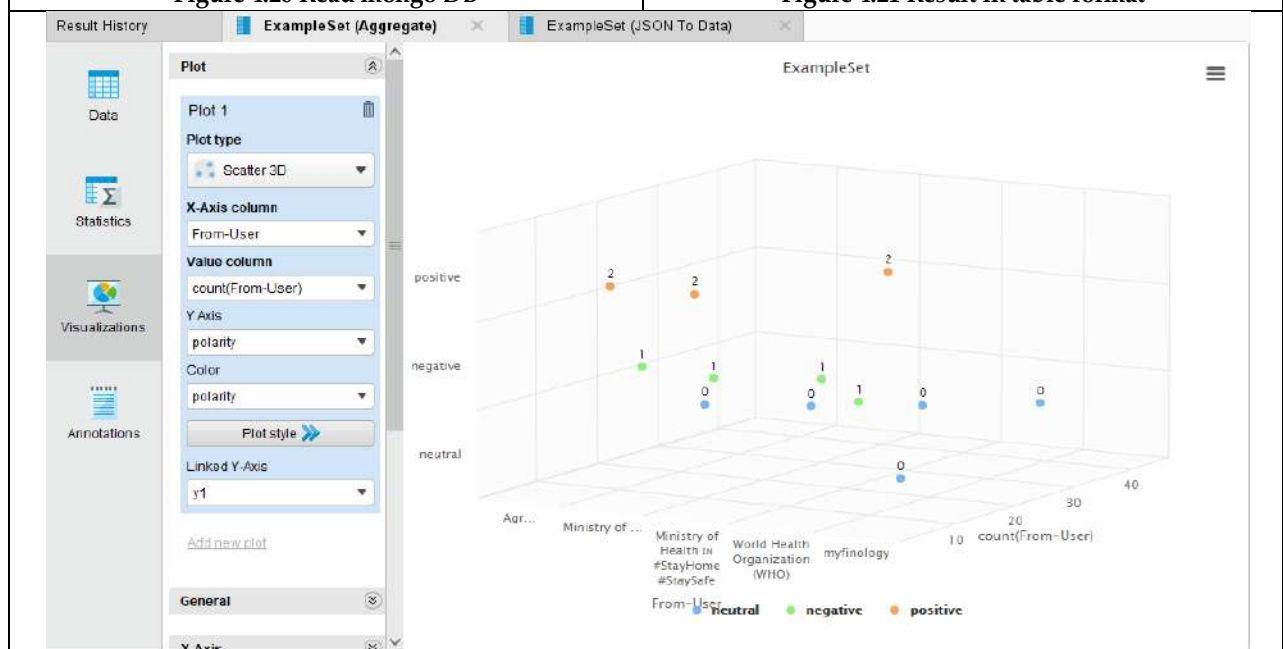
Figure 4.22 Results in pie chart

RESEARCH ARTICLE

Automatic Gender Identification in Unmanned Aerial Surveillance Mechanism

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ABSTRACT

Many real world applications, predominantly the surveillance system, needs gender information for their efficient processing. Most of the available methods use face detection or human dressing pattern identification mechanism to differentiate. Those methods fails miserably when a proper facial image is not received and in places where dressing pattern is similar. Here a better mechanism to determine gender information is elaborated in this paper which can be used in street light cameras and in public monitoring systems. The proposed method captures visible light image and Thermal image. Proficient feature that can store information about gender distinction is extracted from these images. Histogram of Gradient(HoG) and Principle Component Analysis methods are applied in the feature set for optimizing feature set and to get rid of the available noises. The presence of superior feature set makes the proposed method provide accurate result throughout the clock and even during bad weather. Eventually Multi level Local Binary Pattern (MLBP) is implemented and the binary classifier ,SVM is used to effectively differentiate gender. The result of this suggested method is found to be successful and better than other existing methods as this needs as input any casual body image of the person taken even without the knowledge of the person.

Keywords: camera, thermal imaging, live camera, picture, gender identification

INTRODUCTION

Gender inequality and discrimination against women and girls is one of the most pressing concerns in today's society. It hinders their health and well-being. Access to health information and services is rather more difficult for women and girls than among men and boys. Flexibility limitations, a lack of judgement power, lower access to education, discriminatory attitudes among societies and healthcare providers, and an inadequate training and

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awareness of the significant medical problems and requirements of women and girls among healthcare services and health mechanisms are all examples of these barriers. Women too have the right to travel freely and fearlessly in day time as well as in night time. This paper opens a way to safe guard the women community against the physical attack. While travelling alone in any public transport for long distances, there is a constant fear of people's misbehaviour, theft, eve-teasing, or even rape. This research work helps in uplifting the women community by providing them the courage, protection and support they demand at times of difficulty while travelling alone. This research work includes various types of cameras, camera activation, sensors and associated sensor circuitry to detect motion in the vicinity of the implementation. With the help of pictures obtained from visible light and thermal cameras, an algorithm is proposed in this paper to detect the persons' gender. Information about person and the alarming situation's information may be shared to the nearby control room centres to assist the fellow passengers.

Gender identity is a feature that aids in a person's first identification. In humans, there are two sorts of gender separation: male and female. The preferences of different gender groups may differ. Younger boys and girls, for example, may appreciate having toys in their hands, but adults may prefer a variety of clothing styles in a variety of colours and patterns. As a result, defining a group's gender requires familiarity with a wide range of characteristics. Knowing the gender of a community can also help company owners publicise their products as stated by various groups of customers, as well as gather statistically relevant customer information, such as the number of women visiting a retail area or a shopping mall in a given time period. In this real implementation of this research work, we install Live cameras in public places for monitoring the specific purpose of security. The purpose of live cameras is that images from visible and thermal cameras are used to establish body-based gender. It can instantly assess the appearance of the entire body in both visible light and thermal photos, depending on the choices available. The priority characters in the picture's body then offer gender information. The method of gender recognition described below uses a combination of visible light and thermal pictures taken from FLIR data set of the full human body to determine gender. The problem of gender identity has sparked a lot of research. Here, a new gender detection technique that uses a mix of visible light and thermal imaging of the body is used to determine gender.

The advantages using body over face pictures for gender recognition are
 (1) Image Resolution: Low resolution images can help better in gender recognition if the full body image is available.
(2) Focus Change: In practical scenarios, the proper picture of face is hardly received. An overall image of the person is only possible. Also the focus may be the full surroundings and not always be the person.
(3) Acquisition Distance: It is an agreed fact that the camera is placed at some distance away from the person, hence the face photograph won't supply good enough records to gender segregation.
(4) Occlusion. At the point, when the facial part is blocked, the face image probable might not be applied to do away with gender statistics.

State of the Art Review
The majority of earlier research on gender detection systems relied on facial photos with good recognition accuracy [1 -3]. Facial photos, on the other hand, are insufficient for gender detection in surveillance systems given the distance between the examined individual and the equipment. Furthermore, using face photographs for gender detection necessitates the person's consent, which is typically not available in surveillance systems. As a result, the utilisation of human body photographs in surveillance systems can be regarded an alternate technique of gender recognition. Only a few research, however, have looked at how to recognise gender information in a surveillance system that uses photographs of the human body. Cao *et al.* [10] established in their investigation that it is possible to recognise the gender of a person using visible-light images of the human body only.

They employed a boosting strategy to identify the gender of the person using a predesigned feature extraction method, namely the Histogram of Oriented Gradients (HOG) feature, on visible-light photographs of the human body. The results of the experiments on a public database (dubbed the MIT database) including 1000 photos showed that they were able to achieve a recognition accuracy of 75%. To increase recognition performance, Guo *et al.* [11]

used a svm classifier (SVM) and another preconfigured feature method of extraction called biologically inspired characteristics (BIFs) on the same database. They reported that using these methods, the accurate recognition accuracy was increased to 80%, which is greater than the earlier research by Cao *et al.* [10]. The authors employed solely visible-light images for gender recognition in these studies. Because of the vast variety of the human body in visible-light photographs, such as clothing, accessories, backdrop, and other elements, precision is limited. Nguyen *et al.* [5] employed extra gender information collected from thermal pictures of the human body for gender recognition to address this challenge. Thermal images are less affected by variations in clothes or accessories since they are taken based on the difference in temperature between the human body and backdrop regions. As a result, thermal images are more effective at depicting the form of the human body than features on the human body. Nguyen *et al.* [5] used the HOG feature extraction method for feature extraction and SVM for gender recognition on two types of images. The combined picture recognition accuracy was better than utilising only visible-light or solely thermal images for gender recognition, according to their findings. They also employed a unique feature of thermal imaging, in which backdrop regions seem darker than human body regions despite their complex structure, to evaluate the quality of sub-regions in visible and thermal pictures, resulting in a new feature dubbed weighted HOG [12].

This approach is effective at reducing the effects of background regions on the derived HOG features, which improves recognition accuracy. Several additional studies have employed a succession of photographs [13,14] or the three-dimensional (3D) shape of the human body [15,16] for gender recognition in addition to using a single image. The use of predesigned and/or unsuitable feature extraction methods, such as HOG, BIFs, or weighted HOG, limits the accuracy of the approaches discussed earlier in recognising gender information from photos of the human body. Unsuitable extracted features and associated noise might come from the deployment of an ineffective feature extractor, lowering recognition accuracy.

Proposed Method
The various staegs in the proposed methodology for gender identification is narrated in Figure 1.

Human Body Contour Detection.
A strong method for accurately recovering human body outlines from a complicated backdrop environment is used here. Due to changes in the background environment, removing a background scene to separate the human body from the scene is a challenging task. A human detection approach to locate the human body areas in the collected photos during the pre-processing step is necessary . This paper focuses on the characteristics of the human body and how to properly and successfully extract the human body from the background. As a result, the most effective human body extraction process will aid in producing promising results.

Feature Extraction Methods for Gender Recognition
The next step in our suggested strategy is to extract picture features from the photos after detecting body boxes within the images. Using the raw pixels directly is the simplest approach to use these photographs. However, due to the vast variety in these photographs, such as clothing colour, lighting shift, and hairdo, using the raw pixels directly is not a viable solution. We employ two separate feature extraction approaches to extract more discrete gender traits for gender recognition purposes. For inter-experimental comparison, we use the HoG and LBP methodologies to assess the performance of the recognition system.

Histogram of Oriented Gradient
The HoG feature extraction technique has been used to solve problems like human identification [19] and gender recognition [12]. Cao *et al.* [12] used the HoG technique on the issue in a previous studies on gender recognition, and it performed well. As the title suggests, the Histogram of Gradient (HoG) extraction of features method collects image features by capturing the magnitude, weight, and direction of edge characteristics in sub-blocks of an image. As a result, an age map for the entire image can be generated. In our problem, gender identification is largely measured employing body shape and various sorts of edges in a body image. As a result, the HoG feature approach

is efficient for gender recognition, as shown in [12]. In this research, we use the HoG approach to extract visual features for the gender recognition task.

Multi-Level Local Binary Pattern

Lighting variations in the collected body image concerns, as stated in previous sections, add to the challenge of gender recognition based on the human body shots. In earlier computer vision experiments, the local binary pattern (LBP) was shown to be a visually stunning feature extraction method. The main advantage of the LBP is that it is independent by the image's lighting levels. Finger vein recognition [20], face recognition [21], age estimation [16,22,23], gender recognition [24], and face re-identification [25] are only a few of the biometrics systems those have successfully used the LBP. The author suggested adopting a multi-level LBP methodology instead of just a simple LBP for the age estimate challenge in previous work on age estimation estimation [16,22]. By accumulating the histogram of uniform and non-uniform LBP elements, we can recognise features including such lines (edges), corners, and spots in an image. The MLBP feature (image feature extracted using the MLBP technique) has been shown to outperform the LBP feature [16,22]. For this study, we use the MLBP feature extraction method of solving the gender detection problem. The mechanism of LBP is depicted in the described Figure 2.

The radius of a circle within which the surrounding pictures are positioned, as well as the number of surrounding pixels, are important factors in LBP computation. In this presentation, the LBP algorithm is utilized to obtain an illumination invariant characteristics. By comparing the centre pixels with the P bounding box, the LBP feature extraction method acts as an automatic threshold strategy for obtaining the picture shape features. As a result, a P-bit LBP code is used to encode each pixel in a picture. Although changing the image-capturing settings can alter the illumination, the LBP technique maintains the same image attributes independent of illumination. By adjusting the radius and the count of surrounding pixels, we can capture the picture feature at different sized and resolutions.

To construct the picture feature, the retrieved LBP signals are first divided into consistent and quasi codes [16,22]. Consistent codes contain no more than two bit-wise transitions from 0 to 1 in their structure (or 1 to 0). The other forms of codes are described as a non uniform codes. Uniform codes, such as 00000000 and 011111000, contain two bit-wise transitions form 0 to 1 (or 1 to 0), but non-uniform codes, such 01010100 and 01100110, have six as well as four bit-wise transformations, respectively. The uniform codes hold information on the texture's edge, corner, spot, and other features. On the other aspect, non-uniform coding defines exceedingly complex image texture qualities. Noise could possibly be to blame for the diverse textures. Hence, this texture characteristic is insufficient to create an image feature. The orientation of the texture (edge, corner, etc.) is not as significant for the gender recognition problem as the appearance and amount of texture features in the image, as it is for the age estimation problem. Thus, we divide the uniform LBP codes further by evaluating patterns with similar texture shapes but different directions of travel. We make the rotation invariant LBP code [16,22] by completing this stage.

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Implementation Details

The SVM algorithm is used to recognise the gender in the final step of our suggested method. We use the Matlab 2017a package for this. Using support vectors and a kernel function, SVM divides input photos into male and female categories. The SVM is a well-known supervised learning method in machine learning and is mainly used for classification and regression problems using support vectors. Table 2 shows the experimental outcomes employing the HoG and MLBP features. These findings show that combining two different types of photos can improve the recognition system's accuracy. We employed the equal error rate (EER) to evaluate the performance of the gender recognition system, similar to earlier studies on body-based gender recognition [5,12]. The EER is a commonly used main error measurement in recognition systems like finger-vein recognition, iris recognition, and face recognition. The EER is defined as the error that occurs when the false acceptance rate (FAR) equals the false rejection rate (FRR). We have two classes in our gender recognition system: male and female. Hence, "a ground-truth male image that is incorrectly identified as a female image" and "a ground-truth female image that is falsely recognised as a male image" are two conceivable error scenarios. The FAR refers to the first case of error, in which a ground-truth male image is incorrectly recognised as a female image; the FRR refers to the second case of error, in which a ground-truth male image is incorrectly recognised as a female image. In biometrics studies, we usually calculate EER using the genuine acceptance rate (GAR) rather than the FRR number. The GAR is defined as (100 - FRR)/(percent). We always hope that the inaccuracy in the recognition system is as minor as possible. As a result, a lower EER number suggests a superior recognition system. The ultimate recognition accuracy (EER) of the recognition system is calculated by averaging the EERs of fifty images from the testing databases. The results are depicted in table 2.

CONCLUSION & FUTURE ENHANCEMENT

This article discusses the very method of gender determination using a combination of visible and thermal images of the human body through live cameras in unmanned areas in the surveillance system. The recognition accuracy of systems that use a combination of visible image and thermal image to determine gender is discussed. The results are promising and it is targeting a direction towards implementing new schemes for the safety of female gender pedestrians in hazardous situations. It is also suggested that a larger database be compiled for experiments in this area and that recognition indicators be evaluated using different recognition methods. It is also possible to increase recognition accuracy by combating negative effects such as image quality, background, shadow effects, etc

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Table 1 : Details of images taken from the FLIR data set .			
Description of database for our experiments (10 visible-light images/person and 10 corresponding thermal images/person).			
Database	Males	Females	Total
Number of persons	254	158	412 (persons)
Number of images	5080	3160	8240 (images)

Table 2: Analysis of the result achieved in the proposed method.			
SVM Layer Kernel	Recognition System Accuracy Using Combination Visible-Light and Thermal Images		
	Feature-Level Fusion Approach		
	EER	**FAR**	**GAR**
Linear	**12.423**	6	79.015
		10	88.53
		12.44	89.561
		16	93.087
		17	95.78
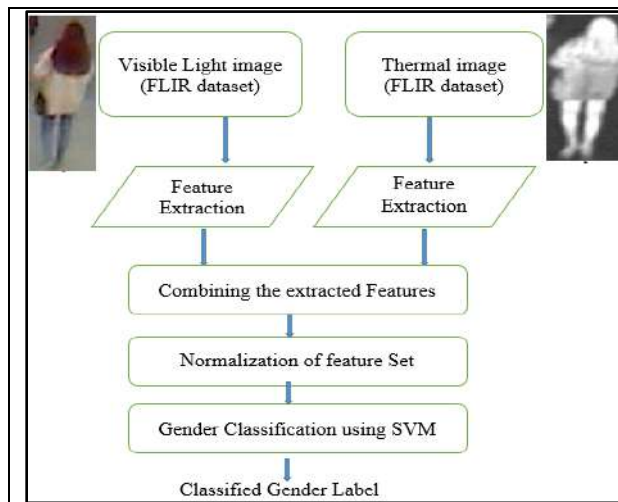
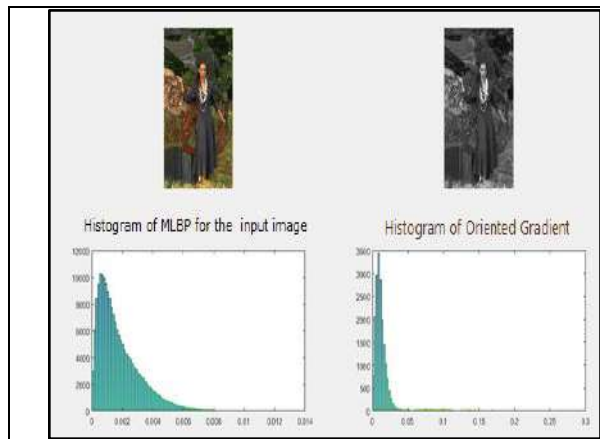|	
Figure1: Stages in the proposed gender identification method	**Figure 2: LBP Method of feature extraction .**
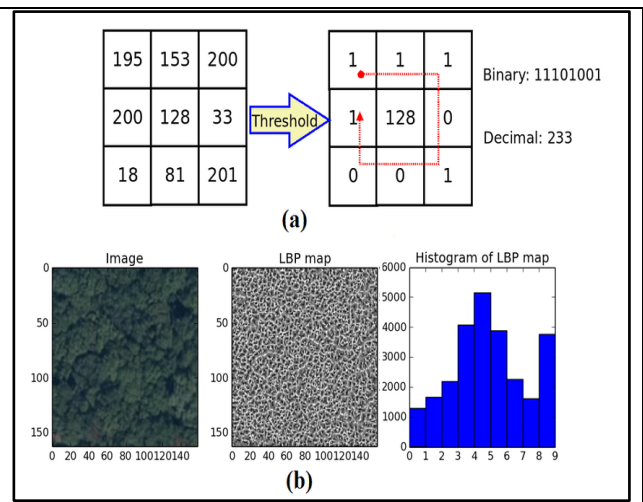
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| Figure 3: Histogram of the MLBP feature and HoG values extracted from the image. | Figure 4: Sample data from FILR containing visible light and thermal images of male and female gender pedestrians |

RESEARCH ARTICLE

On Regular Neutrosophic Fuzzy Matrices

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ABSTRACT

The aim of this paper is introducing the concept of regular Neutrosophic Fuzzy Matrices (NSFM) as a generalization of regular fuzzy matrices. Also, we provide a new representation of Neutrosophic Fuzzy Matrices in terms of its membership, non-membership and indeterminancy fuzzy matrices. Further, the structure of Row space and Column space of an NSFM are obtained. This leads to a characterization of regular NSFM and Invertible NSFM.

Keywords: Fuzzy Matrix, Regular Fuzzy Matrix, Neutrosophic Fuzzy Matrix, Regular Neutrosophic Fuzzy Matrix, Invertible NSFM
MSC Subject Classification: 15B15, 15A09

INTRODUCTION

The fuzzy set (FS) was introduced by L. Zadeh [12] in 1965, where each element had a degree of membership. The neutrosophic set (NS) was introduced by F. Smarandache [6,9,10] in 1999 where each element had three associated defining functions, namely the membership function (T), the non-membership (F) function and the indeterminacy function (I) defined on the universe of discourse X, the three functions are completely independent. Atanassov has introduced and developed the concept of intuitionistic fuzzy sets as a generalization of fuzzy sets [1,2,8].

However, the classical matrix theory sometimes fails to solve the problems involving uncertainties, occurring in an imprecise environment. In [11] Thomason, introduced the fuzzy matrices to represent fuzzy relation in a system based on fuzzy set theory and discussed about the convergence of powers of fuzzy matrix. In 2004, W. B. V. Kandasamy and F. Smarandache introduced fuzzy relational maps and neutrosophic relational maps.

A matrix $A \in F_{mn}$ is said to be regular if there exists $X \in F_{nm}$ such that AXA= A. In this case, X is called age neralized (g-)inverse of A and is denoted by A^-. A {1} denotes the set of all g- inverses of A. In [3,4,5,7] they have developed a theory for Fuzzy Matrices analogous to that for Boolean Matrices [3].

In this paper, we introduce the concept of regular Nuetrosophic Fuzzy Matrices(NSFM) as a generalization of regular Fuzzy Matrices. In section 2, we present the basic definition, notations on NSF M and required results on Regular Fuzzy matrices. In section 3, we provide a new representation of Neutrosophic Fuzzy Matrices in terms of its membership, non-membership and indeterminancy fuzzy matrices. Further, the structure of Row space and Column space of an NSFM are obtained. The regularity of an NSFM is discussed by representing an NSFM as an combination of membership, non-membership and indeterminancy fuzzy matrices and illustrated with suitableexamplesforaregularNSFM.Insection4,the identity NSFM and permutation Neutrosophic Fuzzy Matrices are discussed.

Preliminaries
In this section, some basic definitions and results needed are given. Let $(N)_{mn}$denotesthe setofall mxnNSFMs.

Definition 2.1
A Neutrosophic Fuzzy Matrix (NSFM) of order mxn is defined as A = $(a_{ij})_{mxn}$,where a_{ij} = [a_{ij_μ}, a_{ij_γ}, a_{ij_v}] the ij^{th} element of A is an combination of its membership, non-membership and indeterminancy value. All the elements of an NSFM are combination of its membership, non-membership and indeterminancy value. Let A and B be any two NSFMs. The following operations are defined for any two-element $x \in A$ and $y \in B$, where x=[x_μ,x_γ, x_v]andy=[y_μ,y_γ, y_v]are in [0,1] such that $0 \leq (x_\mu + x_\gamma + x_v) \leq 3$ and $0 \leq y_\mu + y_\gamma + y_v \geq 3$, then

x + y=[max {x_μ,y_μ}, max {x_μ, y_μ}, min{x_v, y_v}]
x .y=[min {x_μ,y_μ}, min{x_μ, y_μ}, max{x_v, y_v}]

Here weshall follow the basic operations on NSFM as given in [5].
ForA=(a_{ij})=[a_{ij_μ},a_{ij_γ}, a_{ij_v}]andB=(b_{ij})=[b_{ij_μ},b_{ij_γ}, b_{ij_v}] of order mxn their sumdenoted as A+B is defined as,

A+ B=(a_{ij} +b_{ij})=[(a_{ij_μ}+b_{ij_μ}),(a_{ij_γ}+ b_{ij_γ}), (a_{ij_v}+ b_{ij_v})]   ….(2.1)
For A= $(a_{ij})_{mxn}$ and B=$(b_{ij})_{nxp}$ their product denoted as AB is defined as,

AB= (c_{ij})=$\sum_{k=1}^{n} a_{jk} . b_{kj}$ i =1, 2, …, m and j=1, 2, …, p
    =$\sum_{k=1}^{n}(a_{ik\mu} . b_{kj\,\mu})$, $\sum_{k=1}^{n}(a_{ik\gamma} . b_{kj\,\gamma})$, $\sum_{k=1}^{n}(a_{ikv} . b_{kj\,v})$i=1,2,…,m and

$$j=1, 2,…,p \quad …..(2.2)$$

Therefore A≤B if and only if $a_{ij_\mu} \leq b_{ij_\mu}$, $a_{ij_\gamma} \leq b_{ij_\gamma}$and $a_{ij_v} \leq b_{ij_v}$ …..(2.3)

In particular if a_{ij_μ}=b_{ij_μ}, a_{ij_γ}=$b_{ij_\gamma}$and a_{ij_v}=b_{ij_v} then(2.2) reduces to the standard max. min composition of Fuzzy Matrices [3,4].
For $A \in N_{mn}$,A^T,A_{i*},A_{*j},R(A),C(A) denotes the transpose of A, i^{th} row of A, j^{th} column of A, row space of A and column space of A respectively. In these quell we shall make use of the following results on Fuzzy Matrices found in [3].

Lemma 2.2
For A,B∈F$_{mn}$, we have the following:

    (i) R(B)⊆R(A)⇔B=XA for some X ∈F$_m$

    (ii) C(B)⊆C(A)⇔B=AY for someY ∈F$_n$

Lemma2.3
For A∈F$_{mn}$,B ∈F$_{np}$, we havethe following:

    (i) R(AB)⊆R(A)and

    (ii) C(AB)⊆ C(B)

Regular Neutrosophic Fuzzy Matrices
In this section, we define regular Neutrosophic Fuzzy Matrices as a generalization of regular Fuzzy Matrices and the structure of Row space, Column space of an NSFM are obtained.

Definition 3.1
For a pair of Fuzzy Matrices C = (c$_{ij}$), D=(d$_{ij}$) and E = (e$_{ij}$) in F$_{mn}$ such that $0 \leq (c_{ij} + d_{ij} + e_{ij}) \leq 3$, let us definethe Neutrosophic Fuzzy Matrix denoted as [C, D, E], whose ijth entry is the combination of c$_{ij}$, d$_{ij}$, and e$_{ij}$that is ([c$_{ij}$,d$_{ij}$, e$_{ij}$]). In particular for C=D=E, NSFM [C, D, E] reduces to C∈F$_{mn}$. ForA=(a$_{ij}$)=([a$_{ij_\mu}$,a$_{ij_\gamma}$, a$_{ij_v}$])∈(NSFM)$_{mn}$, letus define A$_\mu$=(a$_{ij_\mu}$), A$_\gamma$=(a$_{ij_\gamma}$) and A$_v$=(a$_{ij_v}$)

ClearlyA$_\mu$, A$_\gamma$and A$_v$ belongs to F$_{mn}$ such that $0 \leq (A_\mu + A_\gamma + A_v) \leq 3$ and from Definition (3.1) A can be written as A=[A$_\mu$, A$_\gamma$, A$_v$]………(3.1)

Hence forth, weshall use this representation for an NSFM.

Lemma3.2
For A=[A$_\mu$, A$_\gamma$, A$_v$] ∈N$_{mn}$ and B=[B$_\mu$, B$_\gamma$, B$_v$] ∈N$_{np}$, the following hold.

    (i) A^T=[A$_\mu$^{T},A$_\gamma$^{T}, A$_v$^{T}]

    (ii) AB = [A$_\mu$B$_\mu$,A$_\gamma$B$_\gamma$, A$_v$B$_v$]

Proof:
  (i)This directly follows from the Definition(3.1)
  (ii) Let A = [A$_\mu$, A$_\gamma$, A$_v$] = ([a$_{ij_\mu}$, a$_{ij_\gamma}$, a$_{ij_v}$])B=[B$_\mu$, B$_\gamma$, B$_v$]=([B$_{ij_\mu}$,B$_{ij_\gamma}$, B$_{ij_v}$])

Then AB = $\sum_{k=1}^{n}(a_{ik\mu} . b_{kj\,\mu})$, $\sum_{k=1}^{n}(a_{ik\gamma} . b_{kj\,\gamma})$, $\sum_{k=1}^{n}(a_{ikv} . b_{kj\,v})$
i=1,2,…,mandj=1, 2,…,p   (by (2.2))
       =[A$_\mu$B$_\mu$,A$_\gamma$B$_\gamma$, A$_v$B$_v$]
A$_\mu$B$_\mu$ andA$_\gamma$B$_\gamma$ are the max. mincomposition of matrices in F$_{mp}$. Also A$_v$B$_v$is the min max composition of matrices in F$_{mp.}$
Hence the Lemma.

Theorem3.3
LetA= [A$_\mu$, A$_\gamma$, A$_v$]∈N$_{mn}$. Then A is regular NSFM⇔A$_\mu$, A$_\gamma$and A$_v$∈F$_{mn}$ are regular.

Proof
LetA∈N$_{mn}$
If A is regular NSFM, then there exists X ∈N$_{mn}$ such that A X A = A.

Let X=[X_μ, X_γ, X_ν] with X_μ, X_γ, $X_\nu \in F_{nm}$

Then by Lemma(3.2)(ii)

[A_μ, A_γ, A_ν] [X_μ, X_γ, X_ν] [A_μ, A_γ, A_ν] =[A_μ, A_γ, A_ν]

$\Rightarrow$ [$A_\mu X_\mu A_\mu$, $A_\gamma X_\gamma A_\gamma$, $A_\nu X_\nu A_\nu$] = [A_μ, A_γ, A_ν]

$\Rightarrow A_\mu X_\mu A_\mu = A_\mu$, $A_\gamma X_\gamma A_\gamma = A_\gamma$ and $A_\nu X_\nu A_\nu = A_\nu$

$\Rightarrow A_\mu$, A_γ, and A_ν is regular F_{mn}.

Thus, A is regular NSFM$\Leftrightarrow A_\mu$, A_γ and $A_\nu \in F_{mn}$ are regular.

Conversely,

Suppose A_μ, A_γ and A_ν are regular, then $A_\mu X_\mu A_\mu = A_\mu$, $A_\gamma Y_\gamma A_\gamma = A_\gamma$ and $A_\nu Z_\nu A_\nu = A_\nu$ for some X_μ, Y_γ and $Z_\nu \in F_{mn}$. Hence, $X \in A_\mu\{1\}$, $Y_\gamma \in A_\gamma\{1\}$ and $Z_\nu \in A_\nu\{1\}$.

Since $0 \leq (A_\mu + A_\gamma + A_\nu) \leq 3$, it is possible to choose at least one $X_\mu \in A_\mu\{1\}$,

$Y_\gamma \in A_\gamma\{1\}$ and $Z_\nu \in A_\nu\{1\}$ such that, $0 \leq (X_\mu + Y_\gamma + Z_\nu) \leq 3$

Let us define the Nuetrosophic Fuzzy Matrix W=[X_μ, Y_γ, Z_ν]. Then by Lemma (3.2)(ii)

We get, A WA=[A_μ, A_γ, A_ν] [$X_\mu Y_\gamma$, Z_ν] [A_μ, A_γ, A_ν]

= [$A_\mu X_\mu A_\mu$, $A_\gamma Y_\gamma A_\gamma = A_\gamma$, $A_\nu Z_\nu A_\nu$]

= [A_μ, A_γ, A_ν]= A

Thus, A is regular. Hence the theorem.

In Theorem 3.3, A_μ, A_γ and A_ν are to be regular is essential. This is illustrated in the following example.

Example 3.4

Let us consider A =[A_μ, A_γ, A_ν] $\in N_3$ where A_μ= $\begin{pmatrix} 1 & 1 & 0 \\ 0 & 1 & 1 \\ 0 & 0 & 1 \end{pmatrix}$

$A_\gamma = \begin{pmatrix} 1 & 1 & 0 \\ 0 & 1 & 1 \\ 0 & 0 & 1 \end{pmatrix}$

$A_\nu = \begin{pmatrix} 1 & 1 & 0 \\ 1 & 1 & 0 \\ 1 & 1 & 0 \end{pmatrix}$

Then $0 \leq (A_\mu + A_\gamma + A_\nu) \leq 3$

Here A_γ is regular being idempotent and A_μ is not regular, there is no X $\in N_3$ such that, $A_\mu XA_\mu = A_\mu$. Therefore, A is not regular for, if A is regular, then for some X $\in N_3$, AXA= A $\Rightarrow A_\mu X_\mu A_\mu \Rightarrow A_\mu$ is regular which is not possible.

Theorem3.5

Let A=[A_μ, A_γ, A_ν] bean N_{mn}

Then, (i)R(A)=[$R(A_\mu)$,$R(A_\gamma)$, $R(A_\nu)$]$\in N_{1n}$

(ii) C(A)=[$C(A_\mu)$,$C(A_\gamma)$, $C(A_\nu)$)]$\in N_{1m}$

Proof (i) Since A$\circledcirc$ N_{mn}, any vector x$\circledcirc$ R(A)is of the form x=y A for some y$\circledcirc$ N_n that is, x is an combination of membership, non-membership and indeterminancy vector with n components.

Let us compute x $\circledcirc$ R(A)asfollows:

x is aline ar combination of the rows of A$\Rightarrow$ x=$\sum_{i=1}^{m} \alpha_i$. A_{i*} Where A_{i*} is the ith row

of A equating the jth component on both sides yields

$x_j = \sum \alpha_i . a_{ij} \quad i=1 \quad m$

Since, $a_{ij} = [a_{ij_\mu}, a_{ij_\gamma}, a_{ij_\nu}]$

$x_j = \sum \alpha_i . [a_{ij_\mu}, a_{ij_\gamma} a_{ij_\nu}]$ $\quad M \quad i=1$

$= \sum [\alpha_i a_{ij_\mu}, \alpha_i a_{ij_\gamma}, \alpha_i a_{ij_\nu}]$ (By(2.2)) $\quad m \quad i=1$

$= \left(\sum_{i=1}^{m}(\alpha_i . a_{ij_\mu}), \sum_{i=1}^{m}(\alpha_i . a_{ij_\gamma}) \sum^{m}(\alpha_i . a_{ij_\gamma}) \right)$

$= [x_{j_\mu}, x_{j_\gamma}, x_{j_\nu}]$

x_{j_μ} is the jth component of $x_\mu \circledcirc$ R(A_μ), x_{j_γ} is the jth component of $x_\gamma \circledcirc$ R(A)and x_{j_ν} is the jth component of $x_\nu \circledcirc$ R(A) Hence x=[x_μ, x_γ, x_ν]

Therefore, R(A)=[$R(A_\mu)$,$R(A_\gamma)$, $R(A_\nu)$]

(iii)For A = [A_μ, A_γ, A_ν], the transpose of A is A^T = [$A_\mu{}^T$,$A_\gamma{}^T$, $A_\nu{}^T$] By using (i) we get, C(A) = R(A^T) = [$R(A_\mu{}^T)$, $R(A_\gamma{}^T)$, $R(A_\nu{}^T)$] = [$C(A_\mu)$,$C(A_\gamma)$, $C(A_\nu)$].

Hence the theorem.

Theorem3.7

ForA,B $\in N_{mn}$

(i) R(B)$\subseteq$R(A)$\Leftrightarrow$B=XA for some X $\in N_m$

(ii) C(B)$\subseteq$C(A)$\Leftrightarrow$B=AY for some Y $\in N_n$

Proof:

Let A = [A_μ, A_γ, A_ν] and B = [B_μ, B_γ, B_ν]

Since, B=XA,forsomeX $\in N_{mn}$,

Put X = [X_μ, X_γ, X_ν].

Then by Lemma(3.2)(ii),$B_\mu = X_\mu A_\mu B_\gamma = X_\gamma A_\gamma B_\nu = X_\nu A_\nu$

Hence, by Lemma (2.2), R(B_μ) $\subseteq$R(A_μ), R(B_γ)$\subseteq$R(A_γ) and R(B_ν)$\subseteq$R(A_ν)

By Theorem(3.6) (i),R(B)=[$R(B_\mu)$,$R(B_\gamma)$, $R(B_\nu)$]$\subseteq$[$R(A_\mu)$,$R(A_\gamma)$, $R(A_\nu)$]=R(A).

Thus R(B) $\subseteq$ R(A) Conversely, R(B)$\subseteq$R(A)

$\Rightarrow$R(B_μ)$\subseteq$R(A_μ), R(B_γ)$\subseteq$R(A_γ) and R(Bν)$\subseteq$R(A_ν)   (By Theorem(3.6)(i))

$\Rightarrow B_\mu$= XA_μ, B_γ =Y$A_\gamma$and B_ν =ZA_ν and   (By Lemma(2.2))

Then   B=[B$_\mu$, B$_\gamma$, B$_\nu$]
              =[XA$_\mu$, YA$_\gamma$, ZA$_\nu$]
              =[X, Y,Z] [A$_\mu$, A$_\gamma$, A$_\nu$]   (By Lemma(3.2))
              =W [A$_\mu$, A$_\gamma$, A$_\nu$], where X=[X, Y,Z]∈(IVFM)$_{mn}$
              = WAB=WA

Hence the theorem
ii) This can be proved along the samelines as that of (i) and henceomitted.

Theorem3.8
For A∈N$_{mn}$,B∈N$_{np}$, the following hold.
     R(AB)⊆R(A)andC(AB)⊆ C(B)

Proof:
Let A = [A$_\mu$, A$_\gamma$, A$_\nu$] and B = [B$_\mu$, B$_\gamma$, B$_\nu$]
A^T=[A$_\mu$^{T},A$_\gamma$^{T}, A$_\nu$^{T}] and B^T=[B$_\mu$^{T},B$_\gamma$^{T}, B$_\nu$^{T}]
Then by Lemma(3.2)(ii)AB = [A$_\mu$B$_\mu$,A$_\gamma$B$_\gamma$, A$_\nu$B$_\nu$]
By Theorem(3.6) (i)
R(AB)=R([A$_\mu$B$_\mu$,A$_\gamma$B$_\gamma$, A$_\nu$B$_\nu$])=[R(A$_\mu$B$_\mu$),R(A$_\gamma$B$_\gamma$), R(A$_\nu$B$_\nu$)]
⊆[R(A$_\mu$),R(A$_\gamma$), R(A$_\nu$)]=R(A)(By Lemma(2.3))
Therefore, R(AB)⊆R(A)
C(AB) =R((AB)^T)=R(B^{T}A^T)⊆R(B^T)=C(B)C(AB)⊆C (B)
Hence the theorem.

Theorem3.9
For A, B∈N$_n$ with R(A)=R(B)orC(A)=C(B),AisregularNSFM⇔B isregularNSFM.

Proof:
Let A be a regular N$_{mn}$and R(A) = R(B).
Since R(B) ⊆ R(A) by Theorem(3.7) (i),
B = XA = XAA$^-$A = BA$^-$A .
R(A) ⊆ R(B) implies YB = A for some Y ⊚ N$_n$.
Hence B= BA$^-$A = BA$^-$(YB) = B(A$^-$Y)B = BZB. Thus B is regular Converse follows by inter
changing A and B.
A is a regular NSFM⇔B is a NSFM under the condition C(A)=C(B) can be proved along the same lines using
Theorem (3.7)(ii) and hence omitted.
Hence the theorem.

Invertible NSFM
In this section, first we exhibit that the class of all invertible (NSFM) $_n$ coincides with the invertible matrices in F$_n$,
then, as an application, we show that the regularity of NSFM is preserved under congruence relation. In [5], the
identity NSFM is defined as the NSFM in which all the diagonal entries are [1,1, 1] and all other entries are [0,0, 0].
It is denoted by I$_n$. By using the Definition (3.1), the representation (3.1) of the identity NSFM is [I$_n$,I$_n$, I$_n$]=I$_n$.

Definition4.1 Let A∈N$_n$, A is invertible ⇔ AA^T=A^{T}A=Identity NSFM=[I$_n$,I$_n$, I$_n$]=I$_n$.

Definition 4.2 P ∈N$_n$ is a permutation matrix if each row and each column of P contains exactly one interval [1,1, 1]
and all the other entries are [0,0, 0].

Remark 4.3 We observe that P∈N$_n$ is a permutation matrix ⇔PP^T=P^{T}P=I$_n$⇔ P is invertible. Since no two permutation
matrices are comparable, from (3.1) we have, P$_\mu$=P$_\gamma$=P$_\nu$ = P∈F$_n$.

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Theorem 4.4

A is invertible $N_n \Leftrightarrow A$ is invertible in F_n.

Proof:

Let A $=[A_\mu, A_\gamma, A_v]$ then $A^T=[A_\mu{}^T, A_\gamma{}^T, A_v{}^T]$

A is Invertible $\Leftrightarrow AA^T=A^TA$ = Identity N_n.

$\Leftrightarrow [A_\mu, A_\gamma, A_v][A_\mu{}^T, A_\gamma{}^T, A_v{}^T]$ $= [I_n, I_n, I_n]$ = $[A_\mu{}^T, A_\gamma{}^T, A_v{}^T]$ $[A_\mu, A_\gamma, A_v]$

$\Leftrightarrow [A_\mu A_\mu{}^T, A_\gamma A_\gamma{}^T, A_v A_v{}^T] = [I_n, I_n, I_n] = [A_\mu{}^T A_\mu, A_\gamma{}^T A_\gamma, A_v{}^T A_v])$By Lemma 3.2 ii

$\Leftrightarrow A_\mu A_\mu{}^T= I_n, \quad A_\gamma A_\gamma{}^T= I_n \quad$ and $\quad A_v A_v{}^T= I_n$

$\Leftrightarrow A_\mu, A_\gamma$ and $A_v \in F_n$ are invertible.

$\Leftrightarrow A_\mu, A_\gamma$ and A_v are Permutation matrices in F_n. (By [1])

Since $0 \le (A_\mu + A_\gamma + A_v) \le 3$, by Remark (4.3), it follows that $A_\mu = A_\gamma = A_v = A$. Therefore, A is invertible $N_n \Leftrightarrow A$ is a Permutation matrix in F_n. $\Leftrightarrow A$ is invertible in F_n.

Theorem 4.5

Let A $\in N_{mn}$, $P \in N_m$, $Q \in N_n$. A is regular $\Leftrightarrow$ PAQ is regular for permutation matrices P and Q

Proof:

Let A be regular, then there exists X $\in N$ such that AXA = A since P, Q are Permutation matrices by Theorem (4.4) P and Q are invertible. It can be verified that $Q^T XP^T$ is ag-inverse of PAQ. Therefore, PAQ is a regular NSFM. Conversely, Let PAQ be regular then by the preceding part, A = $P^T(PAQ)Q^T$ is regular. Hence the Theorem.

CONCLUSION

The main results in the present paper are the generalization of results on regular Neutrosophic Fuzzymatrices which include the characterization of regular NSFM and preservation of the regularity of NSFM under congruence relation.

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RESEARCH ARTICLE

Analyzing the Knowledge, Attitude and Practice about Obesity among College Students using Python Programming

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ABSTRACT

Obesity is a serious and rapidly progressing condition that causes major morbidity and mortality around the world. The research was based on observational research with a prospective component of weight reduction. For six months, the survey was done with Bengaluru-based college students. According to recent surveys, students are concentrating more on their weight loss. Using python programming data was analyzed the outcome was obtained.

Keywords: obesity, BMI, python programming, knowledge, attitude, practice

INTRODUCTION

Obesity is the most common disease it presents a serious health threat to humans. Obesity is becoming more common not only among adults but also among children and adolescents [1]. The trends in obesity appear to have

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leveled off in recent years, prevention of obesity remains a public health priority due to its high prevalence. The design of obesity varies by sex among racial and ethnic minorities[2]. Obesity raises the threat of a significant number of diseases and ailments, cardiomyopathy, in such, cancer, osteoarthritis, and depression. [3,4] Obesity is caused by a combination of calorie-dense foods consumed in excess and physical inactivity. Overconsumption of calorie-dense foods is one of several factors that contribute to obesity. The catering industry is currently quite successful in mass-producing and marketing calorie-dense meals in both developed and poor countries[5]. Educational achievements, which is allied to socioeconomic status, is also inversely with respect to the percentage of those who are overweight [2]. Obesity is a medical condition whereby an excessive amount of body fat has been collected to the point where it may be harmful to one's health. When a person's body mass index (BMI), which is calculated by dividing a person's weight by the square of their height, exceeds 30 kg/m^2, they are deemed obese, with the range 25–30 kg/m^2 being considered overweight [6].

Genes, endocrine disorders, medications, and mental disorders everyone contributes to a few cases [8] The notion that obese persons consume little and gain weight in the event of slow metabolism is not widely held[9] Obese people consume a fair bit of effort average than non-obese people owing to the energy it is essential to allow due to their bigger body mass. Instead of focusing on cosmetic improvements, weight loss objectives should be realistic and based on evidence of health benefits connected with the intensity at weight loss. A reasonable weight loss goal should be set even as a beginning and realistic objective; the standard recommendation is a 5 % to 10% reduction in overall body mass. Except for serum low-density lipoprotein levels, losing 5% up to 10% of body weight is enough to significantly improve insulin resistance [10,11]. Vikram Rao *et.al.*, (2019); performed an investigation on the incident of obesity and observed it is indeed the case at an all-time high, growing to serve as a solitary of the world's biggest problems. Obesity is related to a variety of health problems, but it furthermore contains a significant impact on the lifestyle, physical ability, mental strength, and healthcare expenses. Obesity happens mostly when body's consumption and production of energy are out of balance. Excess calories are deposited as fat once the quantity of energy consumed exceeds the level of energy expended, resulting in weight increase and, ultimately, obesity.

Rohan Khera *et al.*, (2018); A comprehensive analysis of the literature was conducted for the purpose of uncover randomized clinical trials on the results of FDA-approved weight-loss drugs. Meta-analyses of combinations and systems were performed, with results provided as weighted and discrepancies in standard mean. Bridget Paravattil *et al.*, (2016); conducted a comprehensive conceptual evaluation of topiramate monotherapy among those with type 2 diabetes mellitus who would like to reduce the weight establish its health and effectiveness. Topiramate monotherapy was revealed lower weight in obese included type 2 diabetic individuals. **Rashid Kazerooni, *et al.*, (2016);** has carried out a retrospective investigation to examine if patients taking topiramate for multiple uses experience considerable weight reduction in real-life situations They drew the conclusion that topiramate must be evaluated with higher priority in obesity and being overweight patients for non-weight loss indications due of its low cost, non-controlled status, and efficacy in weight reduction, and that It really should be taken into consideration with higher priority in obesity and being overweight patients for dual benefits.

RESEARCH METHODOLOGY

Study site: This investigation has been conducted in a Bengaluru college.
Study design: This research was implemented in order to use a prospective-observational design.
Study duration: The research was conducted out beyond a six-month period.
Study population and sample: Undergraduate and postgraduate students were described as the target population, with the study sample being 163 people.

Phase of Study

➢ **Phase I:** Baseline period: Determine the statistical sample, including the total number of samples taken. (Pre-test conducted).

➢ **Phase II:** Educating students about obesity with an information leaflet (Post-test conducted).
➢ **Phase III:** Comparing two groups' knowledge, attitudes, and practices are all important factors to consider.
➢ **Phase IV:** Statistical analysis using python programming.
➢ **Phase V:** Result and discussion.

Sources of Data

Students' knowledge, attitudes, and weight-loss habits were often used measure a questionnaire, and their responses to the pretest and post-test were recorded. Students were given weight-loss-related information pamphlets. The student's weight, height, and BMI were measured every two weeks. The trial may have lasted begins with a few weeks to six months. Obesity participants were identified and instructed on how to modify their diets and lives.

Statistical Analysis

Python programming evaluates descriptive data; i.e. for frequency, percentage, chi-square, WS test. Table 1 shows the baseline nature of the research respondents. The study included 163 students (each of categories pre and post-test), and the male-to-female student ratio was 83:80, the mean student age was 21.28830 ± 2.311 years. The predicted mean height was 158.5951 ± 9.0121 including the mean weight was 62.5803 ± 12.2666. The mean BMI was 23.72 ± 3.4 kg/m². From the report, demographic information about age group, the study population's highest age range was identified as 20-22 years (36.8%), the normal age range of the respondents was identified as 23-25 years (30.1%), and the people's lowest age range was identified as 26-28 years (3.7%). The study constituted an almost equal number of male (50.9%) and female (49.1) students. As said in the BMI demographic data, the study population's highest range has been determined to be normal weight students (65.0 %), the average range of the respondents has been determined being overweight students (17.2%), and the least range has been determined to be underweight and obese students (4.9 % and 0%). And these are Figure 1 depicts the situation. To test the association with 'demographic data' and 'pre-test and post-test results' based on knowledge, attitude, and practices regarding obesity; as a consequence the preceding table 3, it's observed that the chi-square value for Post-test knowledge (13.7180) has p has a value of less than 0.05., implying a collateralized between BMI group and post-test knowledge.

Measures Of The Correlation Between Pre-Test And Post-Test Knowledge-Based Questions

With regards to knowledge "BMI is a metric it could be taken to evaluate obesity", majority of students accepted the decision 'definitely' including both the pre-test (68.7%) and post-test (69.9%), for the state Qment "More abdomen weight is riskier than an overall increase in fat distribution in respect of promoting cardiovascular issues", the preponderance of students have accepted the option 'definitely' in the preliminary investigation (39.9%) and post-test (47.9%). Similarly for "Obesity is allied to heart illnesses such as heart attack, blood pressure is high, and so on", the preponderance of students responded 'definitely' in the preliminary investigation (71.8 %) and post-test (78.5 %), "Obesity is allied to diabetes", majority of students responded 'definitely' both in the preliminary investigation (41.7 %) and post-test (41.7 %), for "obesity is linked with osteoarthritis (joint problems)", in pre-test, the preponderance of students identified 'definitely' (41.1%), and in post-test, majority of students identified 'probably' (46.6%), for "fasting/skipping meals is an outstanding technique to drop weight", majority of students identified 'definitely not' in pre-test (54.6%) and post-test (58.3%), for "Excess sugar consumption, whether in the manner of sweets or added sugars in coffee, tea, or milk, is a substantial significant predictor for becoming overweight or obese", the preponderance of students identified 'definitely' in pre-test (46.0%) and 'probably' in post-test (51.5%), for "Sugared beverages (e.g., Pepsi/Coca-Cola) cause weight gain when consumed every now and thereafter." the great majority of students identified 'definitely' in pre-test (47.9%) and post-test (57.7%), for "Frequent eating of fried food causes weight gain", the great majority of students identified 'definitely' in pre-test (65.0%) and post-test (84.7%), for "Excessive eating of refined foods (such as bread, biscuits and momos) causes weight gain", the great majority of students identified 'definitely' in pre-test (45.4%) and post-test (50.3%), for "Stress is a risk factor that accelerates weight gain", 'the great majority of students identified 'probably' in pre-test (33.1%) and post-test (42.3%), for "Regular physical behavior, like as running or participating in sports, is critical for weight loss", the great majority of students identified 'definitely' in pre-test (78.5%) and post-test (82.8%), for "Anti-obesity medicines are indeed the

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most used method of weight loss", the great majority of students identified 'probably' in pre-test (28.8%) and 'definitely not' in post-test (27.6%) and for "Meal complements are good ways to lose weight", the preponderance of students identified 'probably' in pre-test (35.6%) and 'post-test' (38.0). Figure 2 depicts the descriptive metrics of pre- and post-test knowledge-based values.

On evaluating the gap for both pre-test scores and post-test scores on knowledge-based questions by using Wilcoxon's test of signed-rank, it's observed that for the questions: *Obesity is allied to heart illnesses such as heart attack, systolic pressure is increased, and so on; Sugared beverages (Pepsi/Coca-Cola/sweetened juices, etc.) weight gain is caused by when consumed every now and thereafter.; Frequent eating of fried food increases hazard obesity*, the numerological relevance for the WS Rank test is < 0.05. On these knowledge-based questions, a substantial difference between pretest and post-test scores, meaning that a substantial difference between these two pretests and post-test scores.

Measures Of The Correlation With Both Attitude-Based Questions Asked Before/After The Test.

With terms of performance, attitude-based questions, the majority of students accepted the option 'definitely not' in the preliminary investigation (41.7 %) and post-test (53.4 %) for the attitude-based question, "I believe myself to be overweight". With terms of understanding the question, "I believe that my current weight is detrimental to my health", which said that most students including both the pre-test (45.4 %) and post-test (46.6 %) accepted the choice 'definitely not'. Similarly, regarding the question, "I am striving to lose weight", which stated that the preponderance of students have accepted the option 'always' in the preliminary investigation (28.2%) and 'sometimes' in post-test (32.5%), for "I find it challenging to keep my weight steady", in pre-test majority students are identified in 'sometimes' (28.2%) and post-test majority students are identified in 'sometimes' (31.3%), for "Consistent breakfast consumption is something I regard to take part in living a healthy lifestyle", the great majority of students are identified in pre-test is 'definitely' (72.4%) and post-test (82.2%), for "I think small and regular meals help in weight reduction", the great majority of students are identified 'definitely' in pre-test (36.2%) and in post-test (31.3%), for "I am convinced that I would reduce the level of sugar and sweets in my diet", the great majority of students identified 'moderately confident' in pre-test (31.9%) and post-test (37.4%), for "I am certain that I would avoid fried items", the great majority of students identified 'moderately confident' in pre-test (35.6%) and in post-test 'slightly confident'(26.4%), for "In my diet, I'm leaning toward salads and low-calorie snacks over sweets, fried foods, and refined foods.", the great majority of students identified 'very confident' in pre-test (25.2%) and in post-test 'moderately confident' (31.3%) and 'extremely confident' (31.3%), for "I am pleased of my current bodily activity level" the preponderance of students identified 'satisfied' in pre-test (42.3%) and post-test (49.1%), for "I'm ready to engage in active activities like bicycle, outdoor sports, or any other activity that keeps me healthy", the great majority of students identified 'extremely confident' in pre-test (38.7%) and 'very confident' in post-test (32.5%), for "When I have free time, I am planning on doing some form of housework", the great majority of students identified 'very confident' in pre-test (33.1%) and in post-test (38.0%) and "I am certain that I would take the stairs instead of the elevator", the great majority of students identified 'extremely confident' in pre-test (34.4%) and 'very confident' in post-test (33.7%), for "I am certain that I would walk to surrounding locations.", the great majority of students identified 'extremely confident' in pre-test (33.1%) and post-test (38.0%), and "Because I am obese/overweight, I am sad and melancholy", the great majority of students identified never in pre-test (38.0%) and always in post-test (50.3%). The descriptive measures of pre and post-test attitude-based values are represented in figure 3.

On evaluating the similarity among pre-test scores and the consequence of the post-test scores on attitude-based questions by using Wilcoxon's test of signed-rank, it's observed that for the questions: *I'm attempting to slim down; Breakfast intake is something I think to contribute significantly element of living a healthy lifestyle; I am confident that I would reduce my intake of sugars and sweets in my diet; I am prepared to engage in physical activities such as bicycling, participating in other sports, or engaging in any other activity that keeps me healthy.*, it's worth p for WS Rank test is < 0.05, thereby implying whether a significant variance with both pretest scores and post-test scores on these attitude-based questions.

Measures of the Relationship between Pre-Test and Post-Test Practice-Based Questions

With terms of performance, the practice-based question, "I add extra sugars in my coffee/tea/buttermilk", stated that the preponderance of students have accepted the option 'sometimes' in the preliminary investigation (26.4%) and 'rarely' in post-test (33.1%). for "After each meal, I consume a sweet dish.", the preponderance of students have accepted the option 'rarely' in the preliminary investigation (35.0%) and in post-test (41.1%). Similarly, "I use assistants for my domestic activities", which stated that the preponderance of students have accepted the option 'never' in the preliminary investigation (30.1%) and in post-test (40.5%), for "I eat in reaction to stress", the preponderance of students have accepted the option 'some of the times' in the preliminary investigation (35.6%) and in post-test (37.4%), for "I consume beverages that are sweetened with sugar.", both prior to and following the test majority students have identified 'rarely' (56.4%) and (61.3%) respectively, for "I consume fried foods", the great majority of students identified 'rarely' in pre-test is (54.0%) and post-test (63.8%), for "In a week, how often do you eat three main meals and two side dishes?" the great majority of students identified '3-4 times a week' in pre-test (27.0%) and 'once in a week' in post-test (31.3%), for "How many nibbles do you generally consume in a day, aside from its 3 most important meals and two minor meals?", the great majority of students identified 'one day per week' in pre-test (41.1%) and post-test (39.3%), for "I include fruits/salads in my diet", the preponderance of students identified '1-3 times in a week' in pre-test (25.8%) and 'once in 15 days' in post-test (24.5%), for "How regularly do you exercise?" the preponderance of students responded 'every day' in pre-test (24.5%) and 'never' in post-test (32.5%), for "How long do you work out each day?", the great majority of students identified 'not at all' in pre-test (57.7%) and in post-test (57.1%), for "I ask my doctor/dietitian for weight reduction", the preponderance of students identified 'never' in pre-test (62.6%) and in post-test (67.5%), and for "Which among the aforementioned sentences best describes you?", the great majority of students responded 'I currently exercise but not regularly' in pre-test (39.9%) and in post-test (44.2%). The descriptive measures of pre and post-test practice-based values are represented in figure 4.

On evaluating the gap with both pre-test scores and post-test scores on practice-based questions by using Wilcoxon's signed-rank test, it's observed that for the questions; *I use additional sugar in my coffee, tea, and buttermilk.; After each meal, I consume a sweet dish.; How often do you exercise; How long do you work out each day?* it's worth p for WS Rank test is < 0.05, thereby implying there is a great disparity difference between the consequence of the pretest and the results of the post-test on these practice-based questions.

Pre- and Post-Test Scores On Obesity Knowledge, Attitude, And Practice Among Students Descriptive Statistical Analysis Have Been Used To Analyses The Data.

Pre- and post-test scores are utilized to assess students' knowledge, attitude, and practice of obesity, with the findings presented in table 4.

Based on frequency analysis the three chief characteristic attributes are categorized into three types Low, Average, and high and their findings are:

➢ The respondent's scores for pre-test based on knowledge are indeed the foregoing: high (73.6%), average (25.2%), and low (1.2%), and their corresponding scores for post-test was high (85.9%), average (14.1%), and low (0%). On the foundation of these results, It seems to be concluded that the instruction provided following the pre-test is effective and makes significant contributions to students' high-level knowledge.

➢ The respondent's scores for pre-test based on attitude are indeed the foregoing: high (23.9 %) average (75.5 %), and low (0.6%), and their corresponding scores for post-test were high (22.1%), average (76.7%), and low ((1.2%). When the pre-test and post-test attitudes scores are compared, it is clear that the instruction provided following the pre-test is effective and marginally improves the students' moderate-level attitude.

➢ The respondent's scores for pre-test based on practice are indeed the foregoing high (5.5 %), average (81.6 %), and low (0.5 %), and their corresponding scores for post-test was high (0%), average (53.4%), and low (46.6).

Wilcoxon's signed-rank, when comparing two groups, tests were employed overall pre-test and post-test scores based on knowledge, attitude, and practice. Table 5 shows the similarity in between pre-test and post-test scores for the attributes of knowledge and practice; since their p value is < 0.05.

CONCLUSION

Students are very prone to maintain an optimistic outlook toward their general health in the aftermath of the research. They were determined to loose the weight and knew that having a good breakfast was critical to their success. They also made the conscious decision to reduce their sugar intake and were motivated to take part in physical activities such as bicycling, outdoor sports, or any other activity that keeps them fit and healthy. Consequently, education following the pretest aided the student in accurately understanding the overweight or its consequences causes making necessary modifications in their lifestyle attitude and behaviors to avoid obesity.

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Table 1: Illustrates the features of the respondents in the validation phase at the start

Variables	Mean ± S.D.
Age (years)	21.2883 ± 2.311
Gender (Males **:** Females)	83 **:** 80
Height (cm)	158.5951 ± 9.0121
Weight (kg)	62.5803 ± 12.2666
BMI (kg/m^2)	23.72 ± 3.4 kg/m^2

Table 2: Association between statistics on the population and PRE-TEST results based on knowledge, attitude and practices-based questions.

Characteristics		Frequencies (%)	Pre-test Knowledge		Pre-test Attitude		Pre-test Practice	
			Chi square Value	P value	Chi square Value	P value	Chi square Value	P value
Age	17–19	48 (29.4)	8.6510	0.1940	4.5600	0.6010	5.3430	0.5010
	20–22	60 (36.8)						
	23–25	49 (30.1)						
	26–28	6 (3.7)						
Gender	Male	83 (50.9)	5.7310	0.0570	5.9390	0.0510	4.8240	0.0900
	Female	80 (49.1)						
BMI	Underweight	2 (12.9)	5.2060	0.5180	7.0370	0.3170	10.3500	0.1110
	Normal weight	106 (65.0)						
	Over weight	28 (17.2)						
	Obese	8 (4.9)						

Table 3: Association between facts and figures on the demographic and POST-TEST results based on knowledge, attitude and practices-based questions.

Characteristics		Frequencies (%)	Post-test Knowledge		Post-test Attitude		Post-test Practice	
			Chi square Value	p value	Chi square Value	P value	Chi square Value	p value
Age	17–19	48 (29.4)	8.5850	0.0720	5.0170	0.2860	7.9180	0.0950
	20–22	60 (36.8)						
	23–25	49 (30.1)						
	26–28	6 (3.7)						
Gender	Male	83 (50.9)	0.3230	0.8510	3.0690	0.2160	0.1130	0.9450
	Female	80 (49.1)						
BMI	Underweight	2 (12.9)	13.7180	0.0330***	4.5380	0.6040	3.7450	0.7110
	Normal weight	106 (65.0)						
	Over weight	28 (17.2)						
	Obese	8 (4.9)						

Table 4: Contingency table on scores and characteristic attributes

Characteristics		Pre-test Frequencies (%)	Post-test Frequencies (%)
Knowledge	0–49 (Low)	2 (1.2)	0 (0)
	51–74 (Average)	41 (25.2)	23 (14.1)
	75–100 (High)	120 (73.6)	140 (85.9)
Attitude	0–49 (Low)	1 (0.6)	2 (1.2)
	51–74 (Average)	123 (75.5)	125 (76.7)
	75–100 (High)	39 (23.9)	36 (22.1)
Practice	0–49 (Low)	21 (12.9)	76 (46.6)
	51–74 (Average)	133 (81.6)	87 (53.4)
	75–100 (High)	9 (5.5)	0 (0)

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Table 5: Differences in knowledge, attitude, and practice based questions between pre-test and post-test

|		N	Mean Rank	Sum of Ranks	WS Rank test Value	p value
Pre Knowledge – Post Knowledge	Negative Ranks	14	24.5	343	–3.051	0.002***
	Positive Ranks	35	25.2	882		
	Ties	114	–	–		
	Total	163	–	–		
Pre Attitude –Post Attitude	Negative Ranks	31	30.45	944	–0.508	0.611
	Positive Ranks	28	29.5	826		
	Ties	104	–	–		
	Total	163	–	–		
Pre Practice – Post Practice	Negative Ranks	72	43.45	3128.5	–6.39.	0.000***
	Positive Ranks	13	40.5	526.5		
	Ties	78	–	–		
	Total	163	–	–		
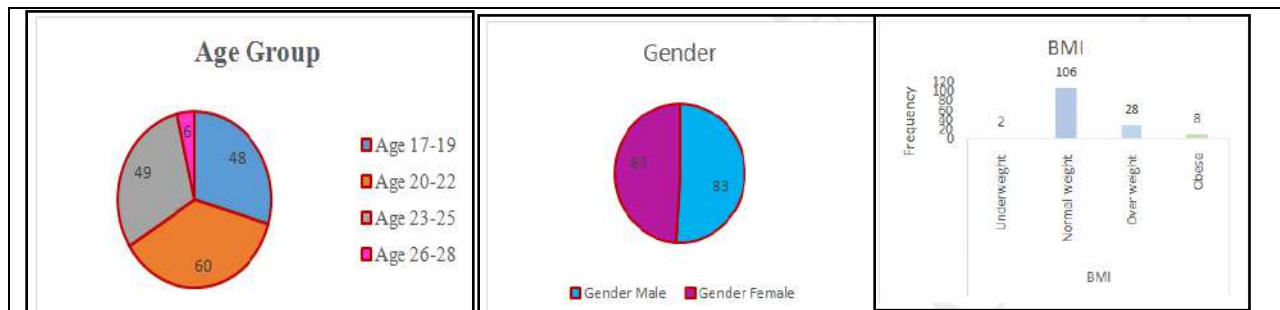
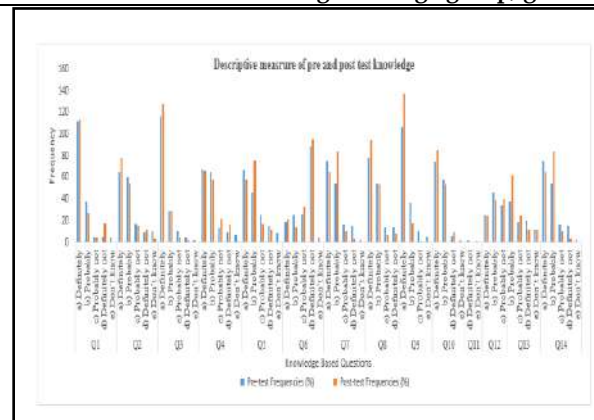**Figure 1: Age group, gender and BMI index demographic data**

Figure 2 – Descriptive measure of pre and post-test knowledge
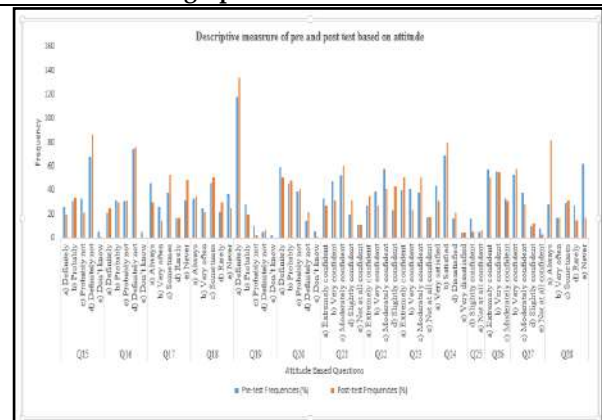

Figure 3: Pre- and post-test attitude descriptive measure
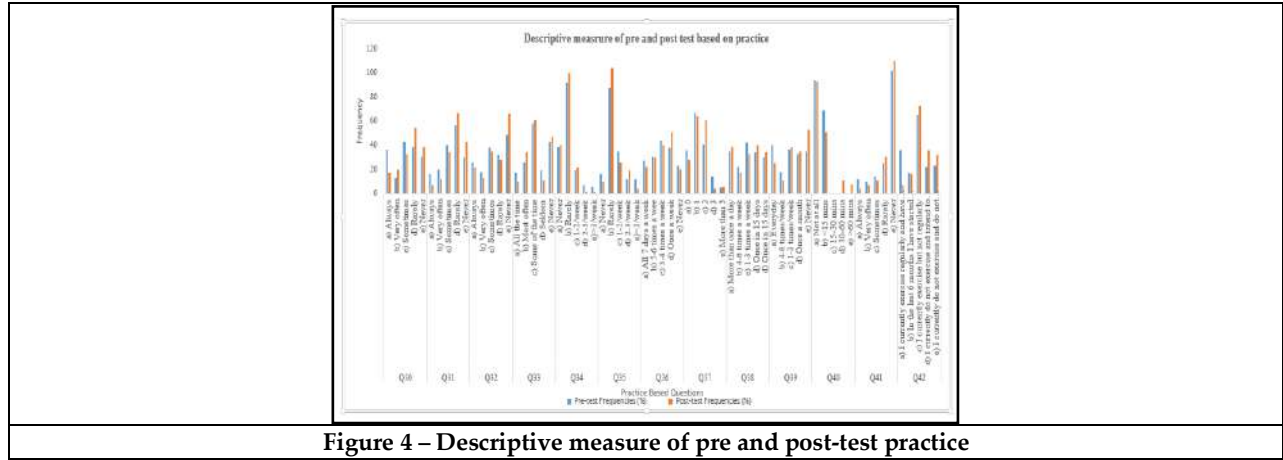
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Figure 4 – Descriptive measure of pre and post-test practice